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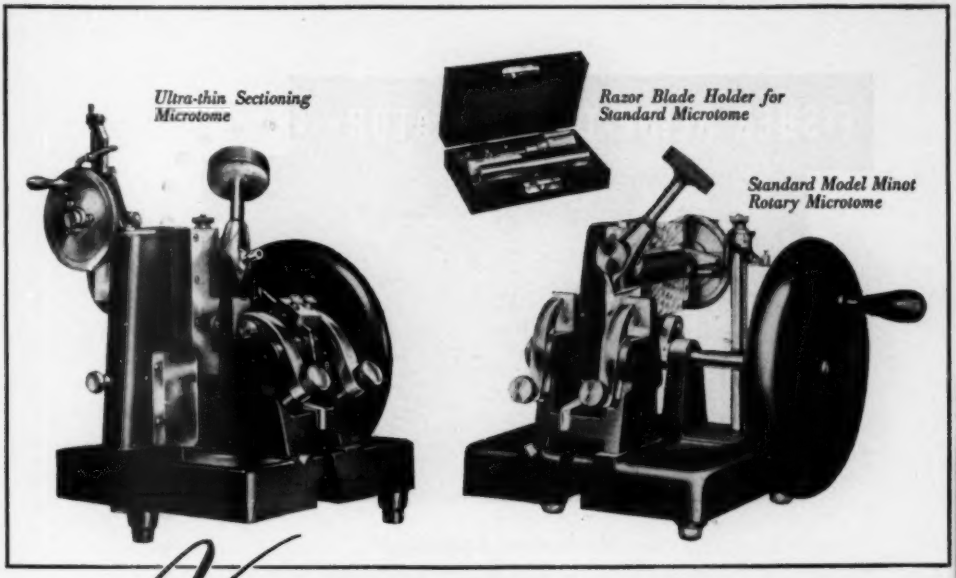

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EXPERIMENTAL MEDULLARY NECROSIS OF THE KIDNEY

A Morphologic and Functional Study

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THE CLINICOPATHOLOGIC entity of necrosis of the renal medulla has only recently begun to be discussed in the American literature.[1] It represents a complication of preexisting urinary tract infection with or without obstruction. However, it is rarely reflected in symptoms sufficiently characteristic to permit a correct diagnosis before the far advanced lesion presents itself at nephrectomy or necropsy. Hence, the pathogenesis of the lesion is not established. It therefore appeared desirable to produce such a lesion in experimental animals at will and without such complicating factors as infection or urinary obstruction, which usually attend the disease in the human. Furthermore, it could be expected that investigation of the functional changes incident to the experimental lesion, especially clearance studies, would contribute to renal physiology.

The lower nephron is considered the seat of selective reabsorption of water and of many solutes in the glomerular filtrate. As early as 1883, Ribbert [2] reported the increased formation of dilute urine in rabbits in which most of the renal medulla had been removed surgically. On the basis of the prevailing theory of renal function, polyuria and impairment of concentrating ability result from damage

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Presented in part at the Thirty-Fourth Annual Meeting of the American Society for Experimental Pathology, in Detroit, April 19, 1949.

From the Communicable Disease Center, Public Health Service, Federal Security Agency, Atlanta, Ga., the Department of Pathology of Northwestern University Medical School, and the Hektoen Institute for Medical Research and the Department of Pathology of Cook County Hospital, Chicago.

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of the lower nephron.[3] Yet in the group of diseases termed by Lucké [4] "lower nephron nephrosis," exhibiting widespread necrotic lesions of the lower tubules, oliguria usually occurs.[5]

Many years ago certain chemicals, such as tetrahydroquinoleine [6] and particularly vinylamine,[7] were shown to produce in animals a medullary necrosis. In order to explore the above-mentioned problems, it appeared promising to investigate the morphologic changes of the different stages of such an experimental lesion and to correlate them with functional alterations as revealed by modern clearance techniques. The vinylamine lesion as produced by Levaditi [7a] in rabbits, rats, mice, guinea pigs, and goats showed medullary necrosis of the coagulation type with shadow-like preservation of the general structure demarcated against the cortical region by leukocytic infiltration and vascular congestion. He observed fibrous replacement of necrotic structures after prolonged survival. Most other attempts at experimental production of medullary necrosis introduced additional complicating injuries, such as ligation of one [8] or both ureters,[1e] combination of one-sided ureteral ligation with retrograde [9] or with intravenous injection of bacteria,[1b] and the intravenous injection of staphylococci in the presence of intact ureters.[10] In some rats on a fat-free diet, necrosis of the renal papilla was observed.[11] However, the most striking lesion was calcium deposition both in the cortical tubular epithelium and in casts. The pyramids—tubules and interstitium—were not directly involved except for their tips. A study of the histologic features of this deficiency lesion uncovers little resemblance to human papillary necrosis.

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MATERIALS AND METHODS

Thirty-six white rabbits, weighing between 2.5 and 4.8 kg., and one dog, weighing 8 kg., received vinylamine. In 26 rabbits and the dog, one or more of the following examinations were carried out on two or more days preceding vinylamine injection and repeatedly thereafter, in

TABLE 1.—*Morphologic and Functional Changes in Animals Intoxicated with Vinylamine*

Prominent Morphologic Findings in Kidneys			Dosage, Ml./Kg.	Days After 1st Injec.	Urine			Plasma, Mg. per 100 Ml.			U/P Ratio	
Cortex	Medulla	Rabbit	× 100		24-Hr. Vol.	Spec. Grav.	Prot.	Creat.	Urea N	N.P.N.	Creat.	Urea
Stage I												
Ischemia	Hyperemia	18A	0.4	1[k]	242	1.012	3+	2.2	54.6	84.9	12.9	6.7
Tubular degeneration	Thrombi	17A	0.4	2[o]	276	1.009	2+	1.3	25.3	44.0	34.3	14.7
		2, 3, 7,	0.8	...	...	.....	...	....	.....	.....	....	....
		8, 14	4.0	...	...	.....	...	....	.....	.....	....	....
Stage II												
± Ischemia	Hyperemia	4	1.3	(1) 1[d]	...	.....	...	1.6	62.0	.....	....	....
Tubular degeneration	Ischemic necrosis	3A	0.25	(1) 2[d]	61	1.011	4+	16.6	188.0	226.7	1.7	0.9
		A	0.7	3[d]	...	.....	...	18.2	.....	224.0	1.3	....
		16A	0.4	4[k]	219†	1.012	+	17.9	288.0	350.0	2.6	1.2
		92	0.3	4[d]	...	.....	...	....	.....	.....	....	....
		B	0.7	4[d]	...	.....	+	9.2	.....	.....	....	....
		Dog	0.6	5[d]	...	.....	...	11.0	.....	206.0	1.1	....
Casts		22	0.5	4[k]	...	.....	...	7.4	.....	167.0	....	....
		52	0.7	5[d]	...	.....	...	....	.....	.....	....	....
	Necrosis	4A	0.25	3[d]	48	1.013	3+	20.2	218.4	330.0	2.9	0.4
	WBC	21	0.5	3[d]	...	.....	...	9.5	.....	185.0	....	....
	Demarcation	14A	0.2	5[d]	...	1.010	3+	21.2	210.0	.....	1.4	0.6
± Ischemia		1A	0.5	(1) 1[d]	...	.....	4+	6.5	67.5	92.4	13.1	3.4
Tubular degeneration		15A	0.4	2[k]	92	1.009	2+	13.1	157.4	.....	2.5	0.8
		2A	0.5	4[d]	35	.....	2+	19.2	285.6	354.4	2.2	0.8
Stage III												
A.												
Diffuse tubular dilatation	Diffuse fibrosis	12A	0.2	(1)32[k]	235*	1.012	+	4.3	77.0	89.2	6.7	4.6
	Circumscribed necrosis	1	0.3-1.5[3]	60[k]	...	.....	3+	....	.....	217.0	....	....
		17A	0.4	23[k]	85	1.015	+	4.4	49.6	71.3	10.0	6.9
		10A	0.1-0.5	73[k]	400†+	1.004	±	7.7	86.9	108.8	4.1	4.7
		5	0.7	19[d]	...	.....	...	....	.....	.....	....	....
	Diffuse fibrosis	7A	0.05-0.3[4]	(9)20[d]	108	1.012	±	1.4	20.3	39.6	51.0	35.8
Fibrosing atrophy		5A	0.05	15[d]	0	.....	...	27.2	280.9	.....	....	....
B.												
Tubular dilatation in patches	Fibrosis in patches	12	0.6	47[k]	...	.....	...	....	.....	.....	....	....
		11A	0.1-1.2[7]	(2)65[d]	71	1.016	±	1.1	18.3	.....	103.1	82.1
		8A	0.05-1.0[6]	76[k]	112	1.011	±	1.9	30.4	37.9	37.6	21.0
		6A	0.05-1.3[9]	(2)97[d]	125	1.015	±	1.4	27.1	40.1	70.5	31.4
± Casts	± Fibrosis	12A	0.4-0.5[2]	25[o]	352	1.009	±	1.2	23.6	41.0	14.1	7.5
			0.6	34[k]	155*	1.009	3+	2.2	50.0	71.6	19.8	8.5
		13	0.6	13[k]	...	.....	...	....	.....	.....	....	....
		9A	0.1	(3)15[k]	275	1.014	±	1.4	17.5	30.9	35.8	32.1
	± Fibrosis	10A	0.1	(2)12[o]	220	1.008	—	1.9	38.8	51.9	29.8	13.6
± Tubular degeneration	Minimal necrosis	15	0.6	7[k]	...	.....	±	1.7	25.0	51.7	....	....
		20	0.8	9[k]	...	.....	+	....	.....	.....	....	....
Range of preinjection values					70-265+	1.024-1.054	— ...	0.8-1.5	13.8-24.0	26.2-49.0	47.5-262.0	20.8-175.0

Key to table: k means killed; o, operated on (left-sided nephrectomy); d, died. Indexes express number of vinylamine injections given (no index means one injection only). A number in parentheses signifies the number of days by which death or nephrectomy followed securing of last functional data. In 24-hr. volume column + signifies that, because of overflow, the total volume was more than noted; † indicates 48-hr., and * 14-hr. volumes.

Note.—Whenever available, the latest functional data obtained before either nephrectomy or death are listed. Of the 36 rabbits studied, three are listed twice (17A, 10A, and 12A), once in referring to results at the time of nephrectomy and once in referring to the results at death.

many instances daily, until the animal either succumbed or was killed (Table 1). Analysis of the urine (collected in flasks containing toluene as preservative) included determinations of the 24-hour volume, the specific gravity, the reaction to litmus paper, and a study of the sediment; furthermore, tests for protein, blood and glucose were made, and the concentrations of creat-

inine,[12] urea nitrogen,[13] and chlorides [14] were determined. In plasma obtained from 5 to 6 ml. of oxalated blood, creatinine,[12] urea,[13] nonprotein nitrogen,[15] and chlorides [14] were measured. No restrictions as to food [16] or water intake were imposed. For histologic study, both 4% formaldehyde solution and Carnoy's solution were used as fixatives. Besides hematoxylin-eosin, stains for hemoglobin (Lepehne), fibrin and neutral fat, as well as Mallory's aniline blue and Gomori's reticulum fiber stains, were studied.

In three rabbits (10A, 12A, and 17A—Table 1) the left kidney was removed surgically under pentobarbital sodium anesthesia two or more days after the first vinylamine injection. These rabbits were followed for varying periods of time.

Three control rabbits were subjected to the same conditions without receiving the poison; no significant morphologic or functional changes developed. In one of them and in six experimental rabbits lesions of spontaneous pyelonephritis were seen. These could be distinguished quite readily from the vinylamine lesions.

Vinylamine,[17] diluted 50 to 100 times with distilled water, was given in doses ranging between 0.0005 and 0.04 ml. per kilogram of body weight. Seven rabbits received it two or more times in the course of three days to three months in progressively increasing doses. The material used presumably contained both vinylamine and its isomer, ethylene-imine, in unknown proportions. The dilutions were prepared immediately before injection because of structural alterations which occur after dilution and render the substance pharmacologically inactive.[7a]

RESULTS

Morphologic Study

Dosage and Survival.—With a dose of less than 0.01 ml. per kilogram of body weight, the period of survival or the degree of renal impairment could not be predicted (Table 1). The difference in individual susceptibility was considerable. Youth seemed to afford protection, while sex played no role. Administration of 0.01 ml. or more per kilogram of weight was invariably fatal within three days and commonly within less than 24 hours (Fig. 1). Death occurred with signs of shock and apparent involvement of the central nervous system as suggested by paralysis of hind legs and convulsions.

The margin between an effective toxic dose permitting prolonged survival and a promptly fatal dose was quite small; e. g., injections of 0.0005 to 0.01 ml. per kilogram created in rabbits 6A and 11A only brief episodes of mild renal impairment and relatively inconspicuous morphologic changes in the kidneys (patchy fibrosis), but 0.013 ml. caused death within a few hours. The range most often resulting in subacute renal impairment was 0.002 to 0.006 ml. per kilogram.

---

12. The method of R. W. Bonsnes and H. H. Taussky, (On the Colorimetric Determination of Creatinine by the Jaffé Reaction, J. Biol. Chem. **158**:581, 1945) was applied in the beginning of this study; thereafter, the modification of it published by R. A. and K. Hare (Determination of Creatinine in Blood and Urine, Federation Proc. **8**:68, 1949; also personal communication to the authors). The difference in results with the two methods as checked in parallel analyses was significant for the purposes of this study.

13. Gentzkow, C. J.: An Accurate Method for the Determination of Blood Urea Nitrogen by Direct Nesslerization, J. Biol. Chem. **143**:531, 1942.

14. Sendroy; Van Slyke, and Hiller, in Hawk, P. B.; Oser, B. L., and Summerson, W. R.: Practical Physiological Chemistry, ed. 12, Philadelphia and Toronto, The Blakiston Company, 1947, p. 575.

15. Koch and McMeekin, in Hawk, Oser, and Summerson,[14] p. 497.

16. The food was purina® rabbit chow checkers, from Rockland Farms, New City, N. Y.

17. Vinylamine was purchased from Bios Laboratories, Inc., New York.

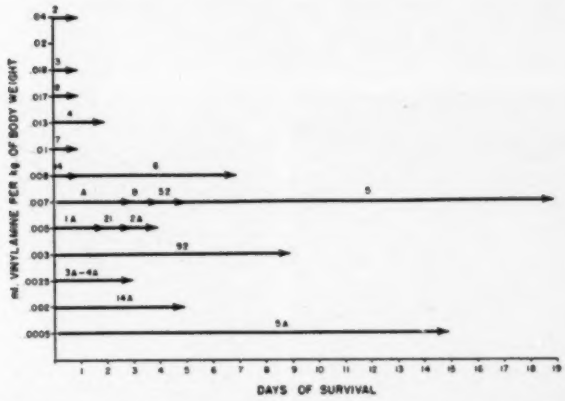

Fig. 1.—Effect of single lethal doses of vinylamine on periods of survival of 19 rabbits. Numbers and letters above arrows refer to individual rabbits which succumbed after receiving a single dose of vinylamine.

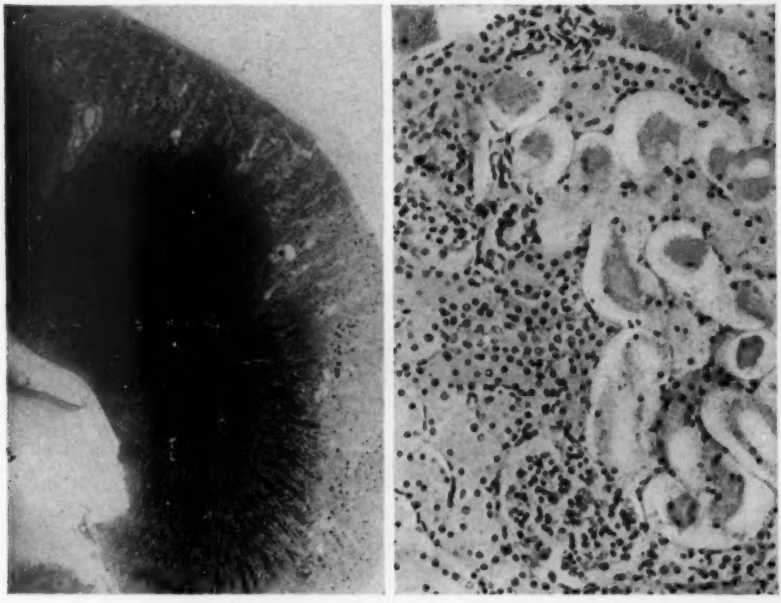

Fig. 2.—Left: Rabbit 14 died within 24 hours after vinylamine injection. Lepehne stain shows excessive hyperemia of medullary intertubular capillaries and of vasa recta bundles at corticomedullary junction. In contrast, there is ischemia of the cortex except for juxtamedullary glomeruli (and larger veins), which are well filled with blood.

Right: Rabbit 3 died within 24 hours after vinylamine injection. The ischemic cortex is seen, with epithelium of proximal tubules showing coagulation necrosis of cytoplasm and lack of nuclear staining. Distal tubules show less injury. A distended vein is seen in the left upper corner. × 200.

Survey of Organs Other Than the Kidneys.—The liver often showed fatty change, hydropic swelling, or central necrosis, usually associated with congestion. The spleen occasionally revealed hyperemia; the lungs, passive congestion and

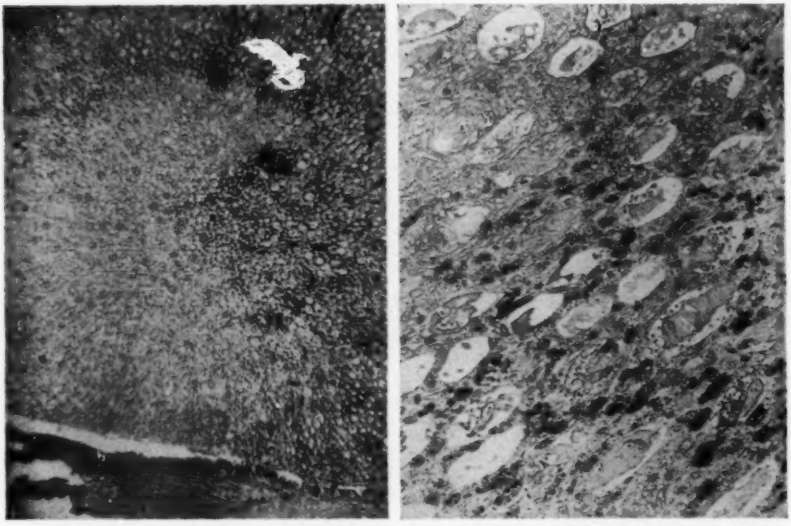

Fig. 3.—Left: Rabbit 15A was killed two days after vinylamine injection. Incipient ischemic necrosis of a renal papilla is shown, with otherwise hyperemic medulla and hyperemic renal pelvis. × 32.

Right: Rabbit 3A died three days after vinylamine injection. Note the medullary necrosis with shadow-like preservation of structure, also protein casts and cellular detritus in Henle's loops and collecting tubules. Many capillaries are engorged with blood cells (early phase of ischemic necrosis or recapillarization?). × 100.

Fig. 4.—Rabbit 4A died three days after vinylamine injection. Necrosis of entire medulla with hyperemic border zone.

edema, circumscribed atelectases or pneumonitis, or both. In two instances, inflammatory foci were encountered in the myocardium. All these changes appeared to be nonspecific. Coccidiosis of the liver was found in six rabbits.

Kidneys: Stage I of Intoxication.—Within the first 24 hours following injection, medullary hyperemia and cortical ischemia appeared (Fig. 2, left), associated with degenerative changes in the convoluted tubules ranging from cloudy swelling to coagulation necrosis (Fig. 2, right). Thus, the histologic picture of this first stage of the intoxication seemed to be compatible with the effect of a shunt of the renal blood flow from the cortex into the medulla as described by Trueta and associates.[18]

Kidneys: Stage II.—In many animals receiving a small enough dose to survive the initial stage, ischemic necrosis developed near the tip of the pyramid (Fig. 3) and often extended to the corticomedullary zone with fairly sharp delineation (Fig. 4). It involved both Henle's loops and collecting tubules, interstitium and blood vessels, with loss of cellular detail but preservation of the general structure

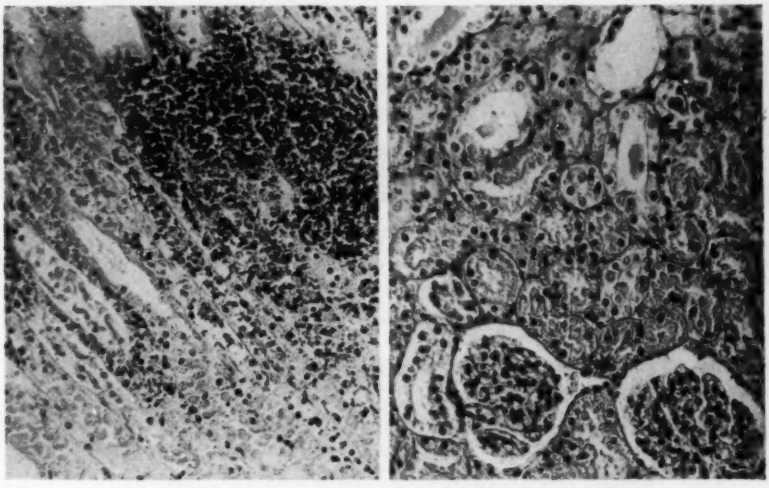

Fig. 5.—Left: Rabbit 4 died two days after vinylamine injection. The corticomedullary junction is seen at the time of early extensive medullary necrosis. Vasa recta show hyperemia and conglutination thrombi. Leukocytic infiltration is seen. There are karyorrhexis and coagulation necrosis of tubular epithelium, protein casts, and edema. × 200.

Right: Rabbit 21 died three days after vinylamine injection. In the presence of extensive medullary necrosis, glomeruli and proximal tubules are practically normal. Distal convolutions contain casts. × 200.

(Fig. 3, right). Demarcation by a zone of engorged and thrombosed blood vessels, edema and hemorrhages was frequently accentuated by an infiltration of segmented leukocytes (Fig. 5, left), becoming more prominent as the border hyperemia subsided. At the same time, the cortex showed in some cases the degenerative changes of the epithelium observed in the first stage, along with regenerative processes and normal blood content of cortical vessels. In other instances the cortex appeared practically normal, except for the presence of casts in distal segments (Fig. 5, right).

18. Trueta, J.; Barclay, A. E.; Daniel, P. M.; Franklin, K. J., and Prichard, M. M. L.: Studies of the Renal Circulation, Springfield, Ill., Charles C Thomas, Publisher, 1947.

Kidneys: Stage III.—In the few observed cases of survival beyond a fully developed Stage II (Rabbits 1, 5A, 7A, and 13A), focal fibroblastic proliferation with subsequent scarring and occasional deposition of lime salts replaced the medullary necrosis (Fig. 6, left). In the cortex, apparently secondary to tubular

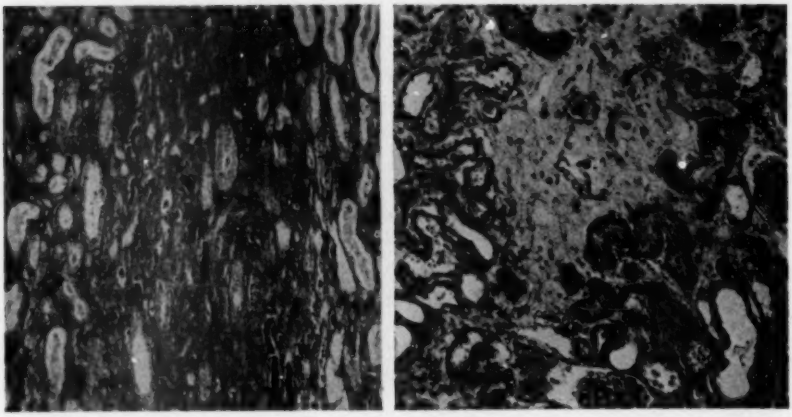

Fig. 6.—Left: Rabbit 1 was killed 60 days after first, and one day after third, vinylamine injection. Note medullary scar with distorted Henle's loops and collecting tubules containing cellular debris and protein casts. Hyperemia and circumscribed hemorrhages were attributed to the last injection. × 100.

Right: Rabbit 13A was killed 33 days after vinylamine injection. Cortex shows fibrotic area with atrophy of glomerulus and tubules. Majority of tubules, however, and capsular spaces are distended because of back pressure. × 100.

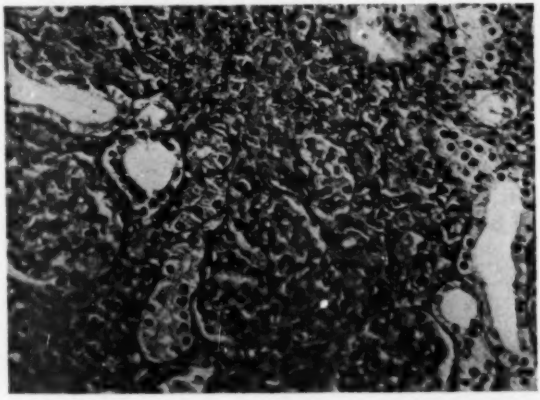

Fig. 7.—Rabbit 5A died 15 days after vinylamine injection. Cortex shows atrophy and obliteration of tubules and reduced capsular spaces, while some distal segments are dilated. × 200.

obstruction caused by the medullary scarring, dilatation of both convoluted tubules and of Bowman's capsules occurred; there were also an increase in connective tissue and atrophy of a few glomeruli and tubules (Fig. 6, right).

In Rabbit 5A, extensive medullary fibrosis without necrosis was associated with marked interstitial infiltration in many cortical areas, with atrophy and obliteration of proximal segments and with glomerular adhesions (Fig. 7)—a lesion unique in the entire series and lacking adequate explanation. This was also the only animal in which the bladder contained yellow gravel, interpreted as inspissated urine.

Poor Reactors and Unilateral Nephrectomy.—The left kidneys of Rabbits 17A and 10A, removed two and 14 days, respectively, after vinylamine injection, showed the expected mild changes: a small amount of papillary necrosis combined in 17A with diffuse medullary hyperemia (early second stage) and in 10A with slight fibrosis and no hyperemia (early third stage). After these rabbits were killed, three weeks and two months later, respectively, the hypertrophic right kidneys revealed extensive medullary fibrosis surrounding a small amount of residual papillary necrosis, in addition to dilatation of most cortical tubules and of glomerular spaces. These changes were more marked in Rabbit 10A, which had a stone in the proximal ureteral orifice. Only minimal alterations, consisting of patchy medullary fibrosis and corresponding tubular dilatation in the cortex, were found in Rabbit 12A, both at nephrectomy and at subsequent killing, as well as in three poor reactors which had not been operated on (6A, 8A, and 11A), all treated with repeated doses over periods of one to three months.

The lower urinary tract exhibited vascular changes similar to those occurring in the renal medulla during Stages I and II. They were particularly noticeable in the renal pelvis (Fig. 3, left), the upper part of the ureter and the bladder, consisting chiefly of distinct edema, hyperemia, and hemorrhages associated with superficial ulcerations.

Functional Study

Urine Output and Analysis.—The urine output was usually suppressed during the first few hours of intoxication, coinciding with shock and the apparent diverting of blood from the cortex into the medulla (Stage I). Therefore, no studies could be made of the urine of the five rabbits that succumbed within a few hours after vinylamine injection (Nos. 2, 3, 7, 8, and 14). The first 24-hour volume, however, was grossly normal and, with the few exceptions indicated below, no significant variations were observed throughout subsequent periods of observation. Marked reduction of urine output in some uremic animals in Stage II could be attributed to their decreased food and water intake (Nos. 4A, 14A, and 2A). Terminal cessation of urine formation was observed in one instance of Stage III only (No. 5A), while in all other rabbits in that stage the output was normal or even above normal (Nos. 13A, 10A, 11A, and 12A). All rabbit urines were alkaline as is the rule in this species.[19]

19. The term "24-hour volume" refers to spontaneously voided urine as collected every morning. Therefore, it reflects very inaccurately the actual quantity of urine produced in each 24-hour period. Hence, only gross changes of urine output could be regarded as significant. For the same reason, urine-plasma (U/P) ratios only, and not clearance values or the quantitative excretion of nitrogenous substances and chlorides, could be utilized. Even U/P ratios can be accepted as approximate values only, since they were calculated on the basis of single blood samples obtained at varying times during the daily urine collection period.

Stage I.—In contrast to the urine volume, the specific gravity showed a consistent and prompt decline on the first day of intoxication, paralleled by a decrease in the urine-plasma (U/P) ratios for creatinine and urea,[20] by marked proteinuria and cylindruria without hematuria or glycosuria, and by increasing azotemia (Figs. 8 to 10). Both plasma levels and U/P ratios of chlorides showed a decrease in the three cases (Nos. 15A, 16A, and 17A) in which they were followed (Fig. 8).[21]

Stage II.—In animals succumbing between the third and the fifth day, i. e., during Stage II (Fig. 10), continued proteinuria and excessive azotemia were accompanied by a depression of the U/P ratio for creatinine far greater than the concomitant drop of the specific gravity of the urine, a depression from a normal range of 47.5-262 to 1.1-2.9, while the specific gravity stabilized itself about 1.010.

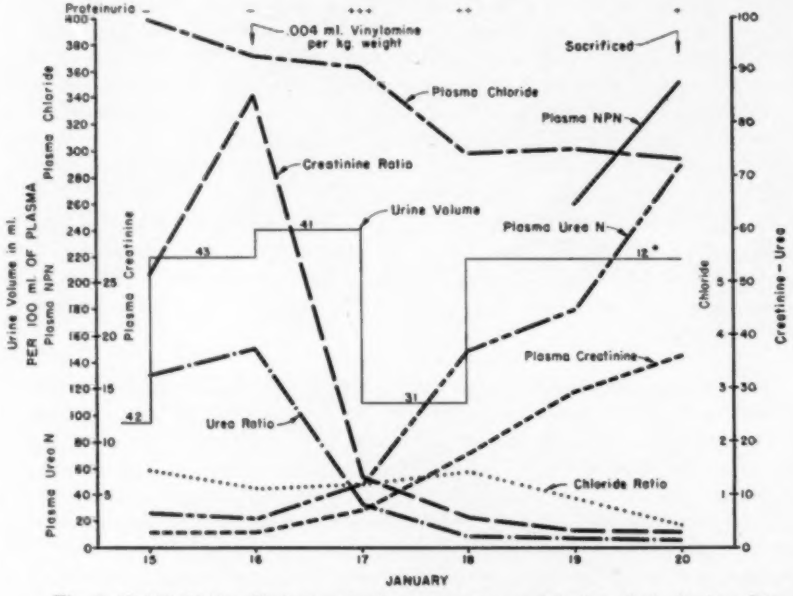

Fig. 8 (Rabbit 16A).—Marked azotemic response to one injection of vinylamine: Rise of creatinine, urea, and total nonprotein nitrogen in plasma, paralleled by a drop in U/P ratio and plasma level of chlorides, by excessive decrease of U/P ratios for creatinine and urea, and by reduction of specific gravity of urine to 1.012. Proteinuria (plus excretion of pus cells and casts) is at first heavy, then lessens. Changes in urine volume are of no apparent significance.

Note.—Specific gravity is indicated in this and following graphs by two-digit figures above the lines signifying urine volume (e. g., 12 instead of 1.012). An asterisk indicates 48-hour volume.

The urea ratio, always below that of creatinine, dropped to 0.4-1.2. Hence, in the presence of complete destruction of the medullary parts of the nephrons, the glomerular filtrate seemed to undergo little concentration on its way down the

20. This reduction in urine-concentrating power could not be estimated adequately, since the first 24-hour collection after vinylamine injection constituted a mixture of urine formed before and after the injection.

21. Other changes not analyzed included progressive anemia and hypoproteinemia.

tubules. The cortical segments also showed evidence of epithelial degeneration in the majority of these cases. However, the animals in which such evidence was lacking (4A and 14A) revealed no greater ability to concentrate urine.

Stage III.—This stage, observed in two rabbits (13A and 5A) which survived a severe uremic response in Stage II, seems to be exemplified by the findings in No. 13A (Fig. 9). These consisted in a moderate though persistent azotemia, slight proteinuria (with some casts and leukocytes in the sediment), polyuria, and hyposthenuria, combined with stabilization of creatinine and urea U/P ratios at levels consistently above those seen in the former group. In contrast, the azotemia in Rabbit 5A (Fig. 10) never leveled off but, enhanced by progressive oliguria and

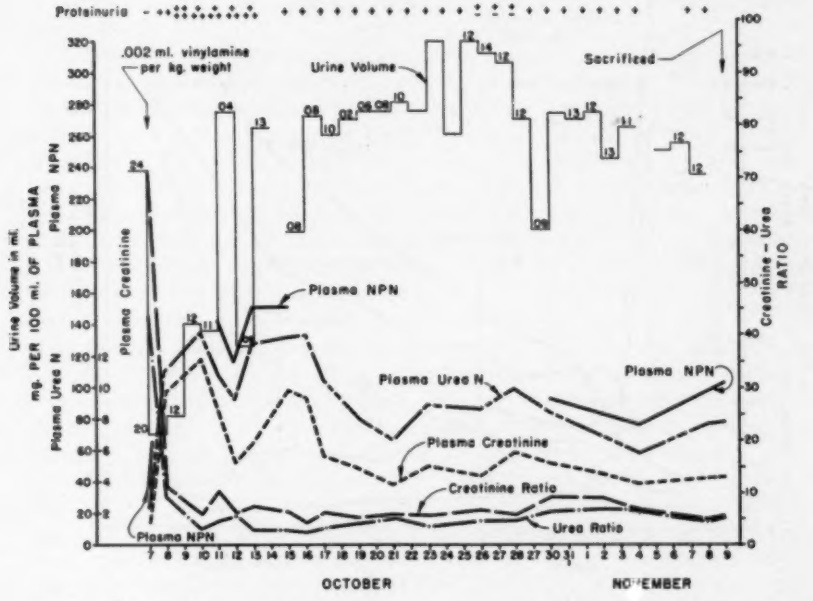

Fig. 9 (Rabbit 13A).—Chronic azotemic response to a single injection of vinylamine: initial rise of nitrogenous substances in plasma (attributed to filtration defect), high plasma level up to the ninth day (produced by combined filtration and reabsorption failure), followed by drop to a moderately elevated plateau (as filtration defect is waning while tubular insufficiency persists). A sharp drop of U/P ratios and of specific gravity of urine occurs initially and persists throughout. Proteinuria, at first heavy, is slight in the chronic stage.

terminal anuria, climbed to the highest value for creatinine encountered in this study, 27 mg. per 100 ml. As another unique finding in our series, this rabbit exhibited, from the third day of intoxication on, microscopic hematuria and, on the third and fifth days, a slightly positive benzidine reaction of the urinary sediment.

Poor Reactors and Unilateral Nephrectomy.—The response in the group of poor reactors (Stage III B) was analogous to that of No. 13A (Fig. 9), though greatly minimized, just as the morphologic features were basically in accord but differed

in extent. Each injection was, as a rule, followed by a more or less well recognizable episode of nitrogen retention and abnormal urinary findings lasting one to a few days. Both functionally and anatomically it appeared that Stage II, medullary necrosis, was skipped and Stage I, the circulatory derangement, either subsided without significant consequences or was promptly succeeded by the fibrous reaction of Stage III. This reaction, developing in scattered patches, was revived by each additional injection but never involved enough nephrons to produce permanent renal failure except when a single kidney had to bear the brunt of metabolic

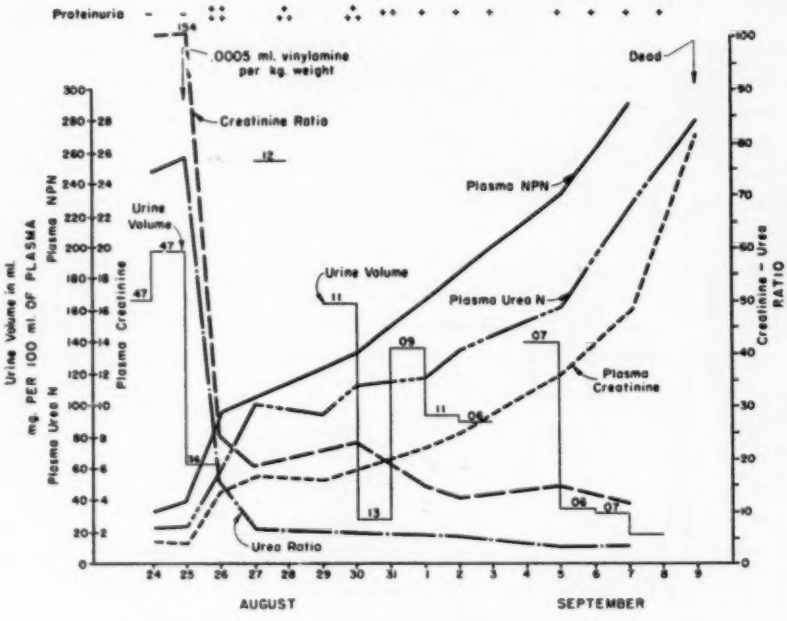

Fig. 10 (Rabbit 5A).—Gradually progressive azotemia follows a single vinylamine injection and reaches excessively high values, enhanced by terminal anuria. Decrease of creatinine and urea U/P ratios is prompt but not excessive, while specific gravity of urine drops to 1.006. Proteinuria, initially heavy, is moderate in the terminal stage.

demands and toxic effect. In two (17A and 10A) of the three rabbits operated on, a slight initial reaction to vinylamine, mirrored by the findings in the surgically removed kidneys, was, soon after nephrectomy, followed by slowly progressive nitrogen retention in the blood and decreasing specific gravity and U/P ratios. This phenomenon, absent in a young rabbit, 12A, was enhanced in No. 10A by two more injections and by intermittent urinary obstruction due to the described calculus.

COMMENT

Morphologic Aspects

Effect of Vinylamine on the Renal Structures.—The effect differs from that of other kidney poisons, such as mercury [22] and uranium,[23] which, as a rule, manifest their main toxic action within a well-defined portion of the nephron, most commonly the proximal segment. A direct protoplasmatic effect of vinylamine as postulated by Levaditi [7a] cannot be excluded. However, it could hardly affect uniformly such diversified structures as Henle's loops, collecting tubules, and the medullary interstitium, on the one hand, and produce a basically different response in the cortex, on the other. For in Stage I the convoluted tubules frequently show their maximal alteration before any epithelial injury in the medulla is visible, while in Stage II, at the time of extensive medullary necrosis, the cortex may reveal no significant abnormality.

The primary effect of vinylamine seems to be neurovascular.[7c] The altered blood distribution, similar to the one described by Trueta and co-workers,[18] could readily explain the cortical alterations: cortical necrosis which is the result of severe ischemia is a well-known clinical entity.[24] The mechanism by which extreme hyperemia in the medulla changes into ischemic necrosis is not so obvious. One feels compelled to invoke as connecting links prolonged vasoparalysis, formation of conglutination thrombi (Ricker's "red stasis" [7c]), and tissue anoxia. Examples of such a process in other organs are central necrosis of the liver in passive congestion and miliary necrosis of the brain caused by multiple vasoparalysis and thrombosis.[25] The marked hemodynamic effect of vinylamine also seems to be attested to by the congestive changes in the lower urinary tract.

Comparison between Experimental and Human Lesions.—The similarity in appearance of Stages II and III and the process observed in reported instances of human papillary necrosis is striking. This, too, exhibits coagulation necrosis of the ischemic type, border hyperemia and thrombotic blood vessels in its periphery, and demarcation by a seam of leukocytes. The necrosis "cuts across all of the main collecting tubules" [24a] and, indeed, across all of the structures of the involved pyramid, producing a ghost-like appearance. Healing may occur by way of fibrosis or by sloughing-off of the necrotic papilla, which may occasionally become the nucleus of, or act like, a renal stone.[26] A similar development of the concrement found in Rabbit 10A is probable. Virulent infection such as is noted in human papillary necrosis may produce vasoparalysis and thrombosis with ensuing interruption of the pyramidal circulation.[1a] The high incidence of human papillary necrosis in diabetes and urinary obstruction may be explained with the well-known favorable effect of these conditions on growth and spread of microorganisms.

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22. (*a*) Richards, A. N.: Direct Observations of Change in Function of the Renal Tubule Caused by Certain Poisons, Tr. A. Am. Physicians **44**:64, 1929. (*b*) Edwards, J. G.: The Renal Tubule (Nephron) as Affected by Mercury, Am. J. Path. **18**:1011, 1942.

23. Hayman and others.[3e] Stoudensky.[8a]

24. (*a*) Sheehan, H. L.: Medullary Necrosis of the Kidneys, Lancet **2**:187, 1937. (*b*) Trueta and others.[18]

25. Scheinker, I. M.: Vasothrombosis of the Central Nervous System: Characteristic Vascular Syndrome Caused by Prolonged State of Vaso-Paralysis, Arch. Neurol. & Psychiat. **53**:171 (March) 1945.

26. Edmondson and others.[1c] Robbins and Angrist.[1d]

FUNCTIONAL ASPECTS

Cortical Defect.—Whether or not a vascular shunt according to Trueta plays any role in human medullary necrosis as intimated by Robbins and Angrist,[1d] or, indeed, whether it exists at all in human physiology or pathology cannot be answered at this time. Even in the rabbit its consistent occurrence has not been agreed on.[27] Nevertheless, it serves at present as the best explanation for the changes seen in Stage I, e. g., the suppression of urine output immediately following vinylamine administration. The resulting glomerular ischemia and reduced filtration pressure would readily account for both the initial proteinuria and the azotemia. The described injury to the proximal segments may enhance proteinuria through diminished absorption of protein which is being filtered in excess of normal through the damaged glomerular membrane.[28] The consistent absence of hematuria and red cell casts in all but Rabbit 5A suggests a minor or transient type of glomerular injury. This is remarkable in the face of the severe medullary destruction. The observed diminution in protein excretion in the later stages may signify either reduced protein filtration because of glomerular recovery or more effective protein resorption because of improved proximal tubular function, or both.

Medullary Defect.—The findings of hyposthenuria and polyuria in Stages II and III evidently reflect the widespread tubular destruction. Normally, the obligatory reabsorption of the glomerular filtrate takes place in the proximal segment, resulting in a fluid devoid of glucose but containing four to five times more creatinine. The final adjustment of the urine concentration (facultative water reabsorption) occurs in the thin limbs and the distal convolutions.[29] A U/P ratio for creatinine of less than 5 has not been observed in normal mammals, even with forced water diuresis.[30] A creatinine ratio of 2 and less associated with azotemia, as seen here, may be due either to reduced passage of creatinine through the glomeruli or to abnormal leakage of creatinine from the tubules or to both. The predominance of tubular injury below the proximal segments points to abnormal back-diffusion as an important cause for the low creatinine U/P ratio, as well as for the azotemia. Indeed, passive reabsorption would seem to be chiefly responsible for the azotemia in Stage III, in which there is little evidence of cortical injury and minimal proteinuria. Stage II probably represents a combination of both types of azotemia with the accent, at first, on faulty filtration, later, on excessive back-diffusion, a combination which may be fatal within five days. A similar though

27. Goodwin, W. E.; Sloan, R. D., and Scott, W. W.: The "Trueta" Renal Vascular "Shunt," J. Urol. **61**:1010, 1949. Reubi, F. C., and Schroeder, H. A.: Can Vascular Shunting be Induced in the Kidney by Vasotropic Drugs? J. Clin. Invest. **28**:114, 1949. Maxwell, M. H.; Breed, E. S., and Smith, H. W.: Significance of the Renal Juxtamedullary Circulation in Man, Am. J. Med. **9**:216, 1950. Kahn, J. R.; Skeggs, L. T., and Shumway, N. P.: Studies of the Renal Circulation, Circulation **1**:445, 1950.

28. Addis, T.: Glomerular Nephritis, New York, The Macmillan Company, 1948.

29. (*a*) Smith, H. W.: Excretion of Water, Bull. New York Acad. Med. **23**:177, 1947. (*b*) Chasis and Smith.[3b]

30. (*a*) Kaplan, B. I., and Smith, H. W.: Excretion of Inulin, Creatinine, Xylose and Urea in the Normal Rabbit, Am. J. Physiol. **113**:354, 1935. (*b*) Shannon, J. A.: Urea Excretion in the Normal Dog During Forced Diuresis, ibid. **122**:782, 1938. (*c*) Ekehorn, G.: Excretion of Urinary Waste-Products Under Abnormal Conditions, with Special Regard to Tubular Function, Acta med. scandinav. **122**:134, 1945; Tubular Function in Renal Deficiency, ibid. **122**:396, 1945; The Reliability of Clearance Tests in Deficient Kidneys, ibid. **123**:269, 1946. (*d*) Chasis and Smith.[3b]

prolonged combination is apparent in Rabbit 5A (Fig. 10), the progressive filtration defect probably contributing to the terminal anuria. Figure 9 illustrates the three phases of azotemia in a single rabbit (13A). The occasional mild and short-lived azotemia in the group of poor reactors may be largely due to filtration disturbance as the immediate result of each new vinylamine injection, the medullary scars being too limited in extent and number to cause overt tubular insufficiency.

The reabsorption factor in the azotemia of some of our rabbits (4A, 14A, 15A, and 2A) is underscored by the finding of a U/P ratio for urea of less than 1. One must assume that urea, normally subject to passive tubular transfer,[31] has, in such instances, escaped from the tubules at a higher rate than even water, so that its concentration becomes lower in the bladder urine than in the plasma. Comparable observations have been made in dogs with acute uranium poisoning [32] and in human cases of "lower nephron nephrosis," [5b] in all of which negative values were encountered for the maximal tubular excretory capacity (*Tm*) as measured with iodopyracet (diodrast®) injection and p-aminohippurate, respectively. Because of such limitations of the clearance methods imposed by tubular injury,[33] computation of the creatinine and other clearances in our series might have been misleading. The degree of renal failure in each experimental animal seems to be best depicted by the plasma creatinine level.[34]

The absence of glucose from all urines speaks for maintenance of some function of the proximal segments. The function of the distal convolutions, however, which includes the facultative reabsorption of water and electrolytes, even if preserved in some of its cells, must have been nullified by the complete destruction of Henle's loops and collecting tubules. This destruction also seems to express itself in the hypochloremia seen in the few rabbits so studied (15A, 16A, and 17A).

Oliguria in "Lower Nephron Nephrosis."—In contrast to the maintenance of urine output in experimental medullary necrosis is the oliguria seen in clinical "lower nephron nephrosis." Passive reabsorption of tubular contents has been proposed as the main cause of this symptom,[35] and this conception has been backed up with results of experimental sublimate poisoning in frogs.[22a] However, such reasoning may be questioned because of differences in urine formation between Amphibia and mammals.[29a] Since mercury affects the tubule almost exclusively in its proximal segment,[36] one must postulate that in mercury poisoning the entire glomerular filtrate is absorbed in that small segment which normally withdraws only about 80 per cent of the filtered fluid. This filtrate, though, is likely to be distinctly reduced in this condition in which glomerular changes occur commonly both in man and in lower mammals,[22b, c] and the cortical blood flow has been found decreased to

31. Kaplan and Smith.[30a] Shannon.[30b] Chasis and Smith.[3b]

32. Bobey, M. E.; Longley, L. P.; Dickes, R.; Price, J. W., and Hayman, J. M., Jr.: The Effect of Uranium Poisoning on Plasma Diodrast Clearance and Renal Plasma Flow in the Dog, Am. J. Physiol. **139**:155, 1943.

33. Popper, H., and Mandel, E. E.: Filtrations- und Resorptionsleistung in der Nierenpathologie, Ergebn. inn. Med. u. Kinderh. **53**:685, 1937. Smith, H. W.: Note on the Interpretation of Clearance Methods in the Diseased Kidney, J. Clin. Invest. **20**:631, 1941. Phillips and Hamilton.[3d] Ekehorn.[30c]

34. Addis.[28] Popper and Mandel.[33]

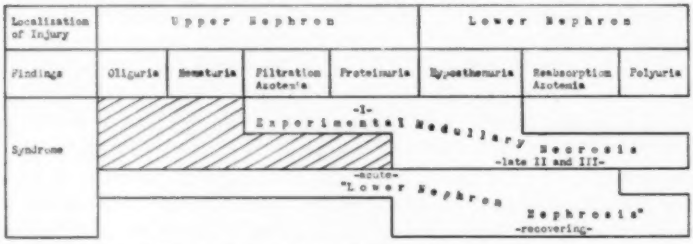
35. Phillips and Hamilton.[3d] Lucké.[4] Moon.[5a] Redish and others.[5b] Marshall and Hoffman.[5c]

36. Kosugi.[7d] Richards.[22a]

40 per cent.[37] Hence, anuria in mercury intoxication should be attributed at least in part to glomerular insufficiency, with the resulting filtrate so small in volume that it may conceivably seep *in toto* out of the tubules before reaching the renal pelvis. Also in "lower nephron nephrosis," pathologic changes of glomeruli have been observed [38]; French [39] used the term "glomerulonephrosis." Cortical ischemia is assumed to play an important role in the genesis of this disease.[3d] Definite proteinuria and hematuria are usual findings. It is therefore hardly justifiable to exclude a glomerular lesion as a significant factor in the lowering or absence of urine output characterizing "lower nephron nephrosis." The well-known polyuria in the recovery phase of this disease suggests restitution of the upper nephron so that the longer-lasting lower nephron injury comes to light,[5b, c] comparable to the shifting of Stage I of medullary necrosis to Stage III.

In view of some of our animals with necrosis of the entire medulla it may be argued that complete passive resorption of tubular fluid was not feasible because of the remoteness of the circulating blood: All pyramidal blood vessels were involved in the necrotic process. However, in rabbits with a lesser lesion, particularly in those in which the process reached Stage III, medullary circulation was, at least in part,

TABLE 2.—*Scheme of Functional Findings in Upper and Lower Nephron Injury*



I, II, and III refer to stages of experimental medullary necrosis.

maintained; yet urine formation never ceased. It required a distinct and progressive cortical injury such as that exhibited in Rabbit 5A to result in terminal anuria.

Lower Nephron Syndrome.—Thus, a correlation of the presented morphologic and functional data appears to furnish new evidence in favor of the conception that renal failure is caused by abnormal tubular reabsorption. While some investigators have been loath to admit such a process,[3b] it can hardly be denied on the basis of animal experimentation [40] or on that of clearance studies in "lower nephron nephrosis" [5b, c] and in other renal diseases.[41] Such an azotemic state, in its pure form, should not be accompanied by hematuria or proteinuria but by polyuria and loss of urine-concentrating power, as postulated in part by other investigators.[42] Hypochloremia may be an additional feature but was not evaluated adequately in this study. Oliguria or anuria and hematuria (barring gross bleeding from open vessels

37. Linder, F., and Sarre, H., quoted from Bell, E. T.: Renal Diseases, Philadelphia, Lea & Febiger, 1946, p. 258.

38. Luckè.[4] Moon.[5a]

39. French, A. J.: Glomerulonephrosis: A Morphological Manifestation of Renal Cortical Ischemia in Toxic Oliguria and Lower Nephron Nephrosis, Arch. Path. **49**:43 (Jan.) 1950.

40. Hayman and others.[3c] Phillips and Hamilton.[3d] Ekehorn.[30c] Bobey and others.[32]

41. Addis.[28] Ekehorn.[30c]

42. Hayman and others.[3c] Phillips and Hamilton.[3d] Addis.[28] Ekehorn.[30c] French.[39]

anywhere in the urinary tract) should be referred to the glomerulus, while proteinuria may be caused by lesions of either the glomeruli or the proximal segments or both. Such a schematic differentiation (Table 2) may well aid in the understanding of clinical problems, although involvement limited to either the upper or the lower nephron hardly ever occurs in spontaneous disease; because of the intimate structural and vascular connections, a combination usually exists, with predominance of one or the other syndrome. This is true of human papillary necrosis, in which the infectious process invariably extends throughout the renal parenchyma, as well as of "lower nephron nephrosis" with its widespread changes involving glomeruli, tubules, and interstitium.

SUMMARY AND CONCLUSIONS

Vinylamine was administered to 36 rabbits and one dog, producing necrosis of the renal medulla in the majority of the animals. This was done in order to (1) elucidate the pathogenesis of human papillary necrosis and (2) investigate alterations in renal function incident to destruction of lower nephrons.

While a direct nephrotoxic effect could not be excluded, the hemodynamic change consistently induced in the kidneys explained adequately the course of the intoxication. This change consisted in a shunt of the renal blood flow, resulting in medullary hyperemia and cortical ischemia, with early degeneration of the epithelium of the convoluted tubules (Stage I). The ischemic medullary necrosis (Stage II) developing thereafter with adequate dosage of the poison was attributed to tissue anoxia caused by excessive medullary hyperemia, vasoparalysis, and multiple thrombus formation, allied with lesser alterations of the cortex. Fibroblastic replacement of the necrotic medulla (Stage III) followed, in the rare cases of survival, with constriction of lower nephrons and secondary dilatation of convoluted tubules.

Morphologic features common to both experimental and human medullary necrosis suggest, as a significant factor in the causation of the latter, circulatory disturbance, i. e., vasoparalysis and thrombosis, precipitated by pyelonephritic infection in the presence of diabetes and/or urinary retention.

Functionally, the transient lesion of the upper nephron (glomerulus and proximal segment) in the initial stage of vinylamine intoxication is reflected in azotemia and proteinuria, complicated by hyposthenuria due to early lower nephron damage. The latter symptom, combined with polyuria and abnormal back-diffusion of nitrogenous substances through inert tubular membranes (as evidenced in a reduction of the urine-plasma ratio of creatinine to almost 1), characterized the late second and the third stage of experimental medullary necrosis. In view of the observation that hematuria and anuria were associated with severe upper nephron atrophy in a single rabbit, a distinction is postulated between "filtration azotemia" and "reabsorption azotemia," the former attended by oliguria and proteinuria with or without hematuria, the latter represented by the findings in Stages II and III, i. e., hyposthenuria and polyuria without hematuria and, theoretically, without proteinuria. Such a distinction may be applied to clinical medicine; thus, in the acute stage of "lower nephron nephrosis," glomerular injury seems to play a prominent role.

The presented findings furnish additional evidence in favor of the lower nephron localization of facultative water reabsorption, without indicating the specific segment.

Experimental vinylamine intoxication is considered a suitable tool for investigations of renal physiology.

PROBLEMS OF CLASSIFICATION OF POLIOMYELITIS VIRUS

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THE CLASSIFICATION of poliomyelitis virus is beset with certain difficulties peculiar to the problem. Obviously, the virus should be named to designate the etiological agent which (1) causes the disease in man and (2) is capable of reproducing the human syndrome in experimental animals. It is unfortunate, however, that Heine-Medin disease, variously known as infantile paralysis or as poliomyelitis acuta anterior, manifests a wide range in the clinical picture and pathological responses it produces in man. As a result, neither the term "infantile" nor "paralysis," nor even "poliomyelitis," truly describes this malady which otherwise exhibits fairly definite epidemiological features. The complexity of this situation is not readily relieved by assuming the existence of corresponding fluctuations in host susceptibility. It is, however, matched by a multiplicity of viral strains which have been recovered, from time to time, in cases diagnosed and reported as poliomyelitis. This is especially true for those strains which, apart from being able to induce infection in monkeys, are primarily or secondarily pathogenic for rodents as well. One is, therefore, left with an impression that what is called Heine-Medin disease may comprise a group of poliomyelitis or poliomyelitis-like disease entities which, in turn, are caused by a group of different yet related viral agents.[1] In a broader sense, such group relationship is further suggested by the fact that spontaneous infections, closely resembling human poliomyelitis on clinical, pathological or epidemiological grounds, are known to occur in at least two other animal species—albino mice and swine. The viruses that cause these diseases (Theiler's virus; Teschen disease virus) are morphologically similar to human poliomyelitis virus but differ with respect to antigenic structure and experimental host range, being strictly monopathogenic for their natural hosts.

Much of present knowledge of human poliomyelitis virus has been derived from studies with rodent-adapted strains. Nevertheless, such strains in frequency of isolation are rare and are far outnumbered by strains that are pathogenic for monkeys only. Hence, continual reference to the classic strains is necessary in order to determine the position of the rodent-pathogenic strains. The experimental approach to the study of "murine poliomyelitis" is marked by two fundamental discoveries: (1) Armstrong's original report [2] that a strain of virus isolated in rhesus monkeys from the medulla of a patient whose disease was fatal could be transferred to mice by intermediary cotton rat passage, and (2) the more recent

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1. Jungeblut, C. W.: J. Pediat. **37**:109, 1950; Helvet. med. acta **17**:167, 1950.

2. Armstrong, C.: Pub. Health Rep. **54**:1719 and 2302, 1939.

demonstration by Dalldorf and Sickles [3] that the feces obtained in many cases of presumable poliomyelitis, abortive or paralytic, yield an agent which selectively paralyzes infant mice. Between the establishment of the Lansing and Coxsackie type of virus, a number of other human strains were reported as having been successfully transferred to rodents. These are known as the Col.Sk virus,[4] the MM virus,[5] the MEF virus,[6] the Y-SK virus [7] and the WW virus.[8] Some of these strains, i. e., MEF, Y-SK and WW, are serologically and biologically closely similar to the Lansing virus, while the Col.SK and MM virus are serologically distinct and have biologic properties which, in some respects, approach those of the Coxsackie virus. While all the above strains were obtained either directly from human poliomyelitic material (cord, blood, feces) or indirectly from monkeys previously infected with such material, two additional neurotropic viruses, which also produce the picture of experimental poliomyelitis or polioencephalomyelitis in rodents, were subsequently isolated from obscure sources, i. e., the virus of so-called encephalomyocarditis (EMC virus) [9] and Mengo encephalomyelitis virus.[10] These two viruses appear biologically and serologically so closely related to Col.SK and MM virus that the four viruses are referred to as Col.SK-MM-EMC-Mengo group.[11] A comprehensive term would be "polioencephalomyelitis group."

There are good reasons for believing that most, if not all, of the above mentioned murine strains are pathogenic for man and capable of producing paralytic or nonparalytic poliomyelitis or polioencephalomyelitis in man. None of them show any immunologic relationship with several strains of Theiler's spontaneous mouse encephalomyelitis, so that the possibility of their having been picked up accidentally in mice is exceedingly remote.[12] Because some of these strains possess certain properties which are divergent from those found with nonadaptable simian strains, much confusion has arisen in the past as to what the criteria should be for the acceptance of a given strain as human poliomyelitis virus, irrespective of the source from which it was originally obtained (human material, sewage, water, flies, rodents, etc.). It is proposed in this paper to discuss this problem in some detail.

EXPERIMENTAL FACTS

I. Discussion of Various Murine Strains

The circumstances under which the various murine strains have been isolated will be briefly reviewed.

3. Dalldorf, G., and Sickles, G. M.: Science **108**:61, 1948.

4. Jungeblut, C. W., and Sanders, M.: J. Exper. Med. **72**:407, 1940.

5. Jungeblut, C. W., and Dalldorf, G.: Am. J. Pub. Health **33**:169, 1943.

6. Schlesinger, R. W.; Morgan, I. M., and Olitsky, P. K.: Science **98**:452, 1943.

7. Melnick, J. L.: J. Immunol. **53**:157, 1946. Lawson, R. B., and Melnick, J. L.: J. Infect. Dis. **80**:201, 1947. Melnick, J. L., and Ward, R: Federation Proc. **7**:308, 1948.

8. Koprowski, H.; Norton, T. W., and McDermott, W.: Pub. Health Rep. **62**:1467, 1947.

9. Schmidt, E. C. H.: Am. J. Path. **24**:97, 1948.

10. Dick, G. W. A.; Smithburn, K. C., and Haddow, A. J.: Brit. J. Exper. Path. **29**:547, 1948. Dick, G. W. A.: Ibid. **29**:559, 1948.

11 (*a*) Dick, G. W. A.: J. Immunol. **62**:375, 1949. (*b*) Warren, J.; Smadel, J. E., and Russ, S. B.: ibid. **62**:387, 1949.

12. Kauffmann, F., and Frantzen, A.: Acta path. et microbiol. scandinav. **25**:356, 1948. Ørskov, J., and Anderson, E. K.: Ibid. **25**:746, 1948.

Lansing, MEF, WW Virus.—The first two strains, i. e., Lansing and MEF, had been isolated from central nervous system tissue in fatal human cases and had received a few preliminary intracerebral passages in rhesus monkeys or baboons. Transfer from the monkeys to mice succeeded only by means of intermediary cotton rat (Sigmodon hispidus hispidus) passage; even though the original simian strains are still available, there is no published record of a successfully repeated adaptation. The third strain, WW, was obtained by inoculating mice directly with blood collected from a paralytic patient during the early stages of poliomyelitis. Again, this experiment has not been repeated. The particle size of murine Lansing virus has been estimated by ultrafiltration as between 8 and 17 millimicrons [13] and by electron microscopy as between 12 and 34 millimicrons,[14] its sedimentation constant as 125 Svedberg units.[15] The murine virus produces typical poliomyelitis in rhesus and in cynomolgus monkeys, even though certain substrains may lose their monkey pathogenicity.[15]

SK Virus.—The most controversial of the various strains of murine poliomyelitis virus is probably Col.SK virus. This virus originated in experiments at Columbia University (1939-1940) in which human SK virus was transferred from early monkey generations (eleventh, fourteenth to sixteenth and eighteenth) to mice by intermediary cotton rat (Sigmodon hispidus littoralis) passage.[4] The human strain had been isolated from feces in a case of abortive poliomyelitis.[16] The observation that the simian SK virus was in later monkey passages pathogenic for rodents was subsequently confirmed by two other laboratories. However, confusing results were obtained in that the respective murine strains showed divergent properties. Thus Toomey (1941-1942), starting with the thirteenth monkey generation of SK virus, obtained on one occasion [17] a low-titered strain, similar to the Lansing virus, whereas another cotton rat adaptation resulted in a strain with high rodent pathogenicity, similar to Col.SK virus.[18] Experiments carried out by Melnick and associates (1946 to 1948)[7] in which the fifteenth, twentieth, twenty-first and twenty-second monkey generations were transmitted to cotton rats yielded only a low-titered murine strain. This strain has since been designated as Yale SK virus.

The paradoxical situation in which two strains, Col.SK and Yale SK, with different biologic and serologic properties, were apparently isolated from the same material requires some comment.[19] A seemingly simple solution has been offered

13. Melnick, J. L.: Bull. New York Acad. Med. **26**:342, 1950.

14. Loring, H. S.; Marton, L., and Schwerdt, C. E.: Proc. Soc. Exper. Biol. & Med. **62**:291, 1946.

15. Theiler, M.: Medicine **20**:443, 1941. Jungeblut, C. W.: Proc. Soc. Exper. Biol. & Med. **72**:532, 1949.

16. Trask, J. D.; Vignec, A. J., and Paul, J. R.: Proc. Soc. Exper. Biol. & Med. **38**:147, 1938.

17. Toomey, J. A., and Takacs, W. S.: Proc. Soc. Exper. Biol. & Med. **47**:123, 1941.

18. Toomey, J. A., and Takacs, W. S.: J. Bact. **43**:87, 1942. Toomey, J. A.: Personal communication to the authors.

19. While viruses, notably bacteriophage, may develop new biologic properties as they adapt themselves to a new tissue environment, they usually preserve their immunologic specificity in that process. However, changes occurring in antigenic structure through mutation or selection of variants, either spontaneously or as the result of animal passage, have been reported in some cases. Examples are influenza virus (Hirst, G. K.: J. Exper. Med. **86**:357, 1947. Briody, B. A.: Bact. Rev. **14**:65, 1950), rabies virus (Wright, J. T., and Habel, K.: J. Immunol. **60**:503, 1948) and foot and mouth disease virus (Michelsen, E., and Mikkelsen, K.: Acta path. et microbiol. scandinav. **22**:406, 1945).

with the suggestion that Col.SK virus was not really adapted from simian SK virus but originated in the wild cotton rats used for the adaptive process.[11a] This concept of a latent cotton rat virus is not in harmony with the fact that the cotton rat, whether infected by central or by peripheral routes, shows extremely high susceptibility to Col.SK virus that existed even on first isolation of the virus. It must also be recalled that at the time these experiments were carried out cotton rats from the same source (Venice, Florida West Coast) were employed on a large scale in attempts to adapt a considerable number of monkey strains, of which the SK strain was only one.[20] None of these strains except the SK strain could be successfully adapted. Adaptation of SK virus could be demonstrated in three successive experiments, yielding in every instance comparable murine strains. Control experiments in which heat-killed SK virus was passed serially through blind cotton rat passages gave completely negative results, providing reasonable assurance that the murine virus had not generated spontaneously in carrier cotton rats. Moreover, spontaneous paralysis has never been observed in wild or laboratory-bred cotton rats,[21] nor has virus been isolated from their feces during an interval of 10 years following the isolation of Col.SK virus. In addition, normal cotton rat serum failed consistently to inactivate the virus not only at the time of the adaptation but again, one year later, in repeated tests carried out with cotton rats supplied from the same source. The absence of antibodies against Col.SK virus in the serum of normal wild-caught cotton rats was once more clearly shown in subsequent work by Warren, Russ and Jeffries.[22] Thus, no experimental proof can be found for this speculation. It is difficult, however, to offer a satisfactory explanation of the facts in the case. One possibility would be that the original SK strain was not a homogeneous virus but represented a mixed virus population containing two variants in an interfering system. Some merit may be found in such a view (1) because human SK virus, on its first isolation in the monkey, exhibited rather unorthodox properties with respect to peripheral infectivity and serologic behavior,[23] (2) because interference between Col.SK and Y-SK virus can readily be demonstrated, resulting in domination of one strain over the other, and (3) because mixtures of Col.SK virus and of an unrelated neurotropic strain, i. e., MEF virus, produce not interference but exaltation.[24] Another assumption would be that the patient from whom the original stool sample was obtained was actually suffering from a double infection with two different viruses. Support for such a hypothesis could be found in the fact that, more recently, Coxsackie virus and classic poliomyelitis virus were encountered together in the stools of a number of patients whose disease had been diagnosed as poliomyelitis. Conceivably, a synthesis of the two types of virus might yield an agent with the properties of Col.SK virus. What

20. Jungeblut, C. W., and Sanders, M.: Proc. Soc. Exper. Biol. & Med. **44**:375, 1940.

21. (*a*) Seegal, B. C.: Proc. Soc. Exper. Biol. & Med. **44**:628, 1940. (*b*) Steinbach, M., and Duca, C. J.: Ibid. **44**:288, 1940. (*c*) Clark, A. R., and Jungeblut, C. W.: J. Nutrition **20**:427, 1940. (*d*) Culbertson, J. T.: J. Parasitol. **27**:45, 1941. (*e*) Culbertson, J. T.; Rose, H. M., and Demarest, C. R.: Am. J. Hyg. **39**:156, 1944.

22. Warren, J.; Russ, S. B., and Jeffries, H.: Proc. Soc. Exper. Biol. & Med. **71**:376, 1949.

23. Trask, J. D.; Paul, J. R., and Vignec, A J.: Proc. Soc. Exper. Biol. & Med. **41**:241, 1939. Vignec, A. J.; Paul, J. R., and Trask, J. D.: Ibid. **41**:246, 1939.

24. Findlay, G. M., and Howard, E. M.: Brit. J. Exper. Path. **31**:45, 1950.

the correct answer may be will probably remain undecided for some time to come.[25] Col.SK murine virus has been estimated to have a particle size between 9 and 14 millimicrons by ultrafiltration [26] and between 25 and 30 millimicrons by electron microscopy,[27] with a sedimentation constant of 130 Svedberg units.[28] The virus produces in rhesus monkeys rarely more than subclinical infection [29] but is highly pathogenic for cynomolgus and African green (Cercopithecus) monkeys, inducing in these animals typical poliomyelitis.[30]

MM Virus.—This strain was isolated by direct inoculation of hamsters with medulla from a fatal case of bulbar poliomyelitis. The original material also paralyzed rhesus monkeys, but the simian virus could not be maintained in serial monkey subpassages. The murine strain, on the other hand, proved readily transmissible in further rodent passages, including hamsters and mice. The particle size of murine MM virus has been estimated at 11 to 16 millimicrons by ultrafiltration [31] and between 10 and 20 millimicrons by electron microscopy,[32] with a sedimentation

25. In a recent publication Melnick (Bact. Rev. **14**:233, 1950) has further simplified the matter, not only by assuming that Col.SK virus originated in "Florida rats" (presumably cotton rats) but also by suggesting that MM virus originated in "New York hamsters" or else was picked up at the Columbia laboratory. In support of this assumption, he quotes the discovery of EMC antibodies, by Warren and associates,[22] in the serums of five wild rats (Rattus norvegicus) trapped in Dania, Fla., where EMC virus was isolated. The experimental evidence that speaks against the cotton rat origin of Col.SK virus has been fully discussed above. It remains to point out that Florida rats (Rattus norvegicus) and Florida cotton rats (Sigmodon hispidus littoralis) are two entirely different species, the first highly resistant and the latter highly susceptible to Col.SK virus. Hence, the two kinds of rodents cannot possibly be expected to play a similar epidemiological role as potential latent carriers of Col.SK or EMC virus. This distinction was also quite clear to Warren and associates [22] at the time of their discovery. Whatever coincidence, therefore, there seems to be between the two sets of observations appears to be one of chance. However, this does not exclude a possibility that viruses of the Col.SK group may have an active reservoir in certain species of resistant wild rodents, of which the common wild rat (Rattus norvegicus; Rattus alexandrinus) is, perhaps the only one known at this time. The subject is more fully discussed in the section on EMC virus.

According to a recent report by Koch (Ztschr. Kinderh. **68**:138, 1950) a virus of the Col. SK-MM-EMC type has been isolated in Germany from four of seven patients with an obscure illness (abortive poliomyelitis or aseptic meningitis). In one patient who died interstitial myocarditis was noted at autopsy; the central nervous system was not examined. The virus was obtained by transfer of blood, spinal fluid, stools and other materials to hamsters and mice. Antibodies neutralizing this virus, as well as MM virus, were demonstrated in the serums of four patients, including one from whom the virus had been obtained. Cross neutralization tests with immune serums against the German F virus, MM virus and EMC virus indicated that all viral strains were antigenically closely related or were identical. Reports of similar virus isolation in Germany have since been published by Bieling (Tr. Austrian Soc. Microbiol., Salzburg, 1950) and by Beller and Keller (Klin. Wchnschr. **27**:422, 1949).

26. Sanders, M., and Jungeblut, C. W.: J. Exper. Med. **75**:631, 1942.

27. Jungeblut, C. W., and Bourdillon, J.: Electronmicrography of Murine Poliomyelitis Virus Preparations, J. A. M. A. **123**:399 (Oct. 16) 1943. Loring, H. S.: Proc. Soc. Exper. Biol. & Med. **64**:101, 1947.

28. Bourdillon, J.: Arch. Biochem. **3**:285, 1944.

29. Jungeblut, C. W.; Sanders, M., and Feiner, R. R.: J. Exper. Med. **75**:611, 1942.

30. (*a*) Jungeblut, C. W.: Proc. Soc. Exper. Biol. & Med. **72**:534, 1949. (*b*) Jungeblut, C. W.: Bull. New York Acad. Med. **26**:571, 1950. (*c*) Verlinde, J. D.; De Baan, P., and Vercruysse, J. A.: Antonie van Leeuwenhoek **16**:9, 1950.

31. Quigley, J. J.: Proc. Soc. Exper. Biol. & Med. **72**:434, 1949.

32. Gollan, F., and Marvin, J. F.: Proc. Soc. Exper. Biol. & Med. **67**:366, 1948.

constant corresponding to that of Lansing and Theiler's FA virus.[33] Murine MM virus, as a rule, produces only subclinical infection in rhesus monkeys but induces typical poliomyelitis in cynomolgus monkeys.[30a]

EMC Virus.[9]—Between November 1944 and October 1945 there occurred three unexplained deaths among one gibbon and two chimpanzees on exhibit at the Anthropoid Ape Research Station at Dania, Florida East Coast. All three animals had been in close contact with visitors. Death occurred unexpectedly and suddenly, or after only short illness, with symptoms of cardiovascular collapse. In all three cases the autopsy showed massive myocarditis and marked pulmonary edema but no pathological changes in the central nervous system. In the first case no attempt was made to recover the etiological agent. In the second case the chest fluid and a saline suspension of ground spleen were injected intracerebrally or intraperitoneally into mice, and the virus was thus recovered. In the third case, mice inoculated with pleural fluid failed to present any of the typical lesions. In mice into which the infectious specimen had been injected, flaccid paralysis developed and, also, provided death did not intervene too soon, extensive involvement of the heart muscle with necrosis of fibers and cellular infiltration. Hamsters and guinea pigs were also susceptible to the virus and showed similar symptoms. Sections through the spinal cords of paralyzed mice showed extensive destruction of all the outer neural elements, but the ganglion cells were apparently not involved by this myelitis. Foci of encephalitis could be seen in the brain, with perivascular cuffing of adjacent vessels; however, no encephalitis was encountered in the guinea pigs. The diameter of the virus, as determined by electron microscopy, has been estimated to be approximately 30 to 35 millimicrons, with a sedimentation constant of about 153 Svedberg units.[34] Murine EMC virus was subsequently shown to be highly pathogenic for cynomolgus monkeys, producing flaccid paralysis with typical poliomyelitic lesions in the spinal cord.[30a]

The source of infection still remains obscure, even though the original observers emphasized the intimate contact with man. That typical poliomyelitis may develop in chimpanzees in natural contact with human patients or carriers is amply documented by experience elsewhere.[35] An epidemiological survey of the incidence of poliomyelitis in Florida between 1944 and 1946 (fig. 1) fails to reveal any immediate and convincing temporal or local relationships with reported human cases; however, it does not take into consideration a multitude of potential carriers among Northern transients, who presumably constituted the greater part of the visitors at the station. Another and different clue is offered through the work of Warren and associates,[22] who could demonstrate the presence of specific virucidal antibodies in the serum of five wild rats (Rattus norvegicus) caught in the town of Dania in 1949. This observation suggests a local reservoir of the virus in wild rats. It is not clear, however, whether the anthropoid apes received the virus from the rats (possibly through insect bite) or the rats from the anthropoid apes (possibly through fecal contamination), and the point may be argued either way. In view of

33. Gard, S., in Proceedings of the Fourth International Congress for Microbiology, 1947, Copenhagen, Rosenkilde & Bagger, 1949.

34. (*a*) Weil, M. L.; Warren, J.; Russ, S. B.; Breese, S. S., and Jeffries, H.: Bacteriology Proceedings, 1950, p. 83. (*b*) Warren, J.: Bact. Rev. **14**:200, 1950.

35. Müller, W.: Monatsschr. f. Kinderh. **63**:134, 1935.

the time element, though, it seems not impossible that the outbreak among the apes may have preceded chronologically the carrier state in the rats. This is, of course, a rank speculation, but the inference is in conformity with the general epidemiological features observed in a nation-wide survey of the same problem. While a very high percentage of neutralizing rat serums was found among samples from certain areas, samples of similar size from adjacent areas gave either completely negative returns or indicated a much lower incidence of positive serums. This sharp dependence on local environment is quite different from the almost universal dissemination of Theiler's virus among albino mice, in which the disease seems to have been established over a long period of time. By contrast, it suggests that EMC virus has been introduced rather recently into rat populations under the controlling influence of regionally operating factors. In the absence of other explanations it is not inconceivable that the source of infection may ultimately be found in man.

Mengo Virus.[10]—This virus is of special interest because there is direct evidence that, apart from being pathogenic for laboratory animals, it is also pathogenic for

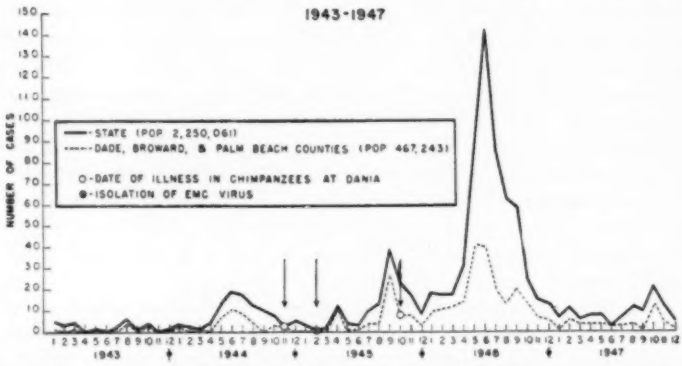

Fig. 1.—Cases of poliomyelitis in Florida from 1943 to 1947. The chart shows the statewide distribution of cases and the cases reported from the general location of Dania.

man.[36] The agent was originally isolated from a spontaneously paralyzed rhesus monkey which had been captive in the outside monkey quarters of the Yellow Fever Research Institute in Entebbe, Uganda. Subsequently, the same virus was isolated from mosquitoes captured in the same locality. Finally, there was also reported a case of human infection with this virus, in one of the laboratory workers. The clinical history of the patient refers to symptoms of mild encephalomyelitis (headache, irritability, delirium, deafness), but symptoms of transient paralysis were also present, involving the muscles of deglutition and the upper fibers of the trapezius muscle. Had the case occurred in the midst of a poliomyelitis epidemic, it would probably have been listed clinically as a case of polioencephalomyelitis, with a presumable localization of the lesion in the upper cervical region of the cord and involvement of the nuclei of the eighth, ninth and eleventh cranial nerves. The virus isolated from the patient's blood during the acute stage produced fever or muscle weakness in rhesus monkeys and fatal encephalomyelitis in mice and in guinea pigs.

36. Dick, G. W. A.; Best, A. M.; Haddow, A. J., and Smithburn, K. C.: Lancet **2**:286, 1948.

The identity of the human strain was established through neutralization tests with the patient's serum, on the one hand, and the original Mengo virus, on the other. The exact size of Mengo virus has not been determined, except that it is a very small, filter-passing agent.

Experimental studies with Mengo virus indicate that the agent, even though originally isolated from a rhesus monkey with flaccid paralysis, possesses more virulence for rodents than for monkeys. Thus, the virus could be readily passed in mice and in guinea pigs, but difficulties arose in passing it from rhesus to rhesus. Flaccid paralysis, however, with central nervous system lesions was obtained in African baboons (Cercopithecus aethiops). Invariably the lesions were more prominent in the anterior horn of the spinal cord than in the brain; also, cord tissue contained more transmissible virus than brain tissue. The higher susceptibility of monkeys, other than rhesus, for Mengo virus appears to be typical of the entire group of viruses, the same phenomenon having been observed with Col.SK virus, which is definitely more pathogenic for cynomolgus (Java) monkeys than for rhesus monkeys.[30a] The reasons for such varying reactivity among different species of monkeys are obscure. Possibly the rhesus monkey responds selectively to the neurotropic qualities of poliomyelitis virus, whereas the cynomolgus monkey registers the viscerotropic qualities of the virus as well.[37] This might also explain why cynomolgus monkeys are used more successfully in isolating poliomyelitis virus from extrahuman sources, such as flies.[38]

Coxsackie Virus.—Isolation of the Coxsackie type of virus is still too recent to permit any final conclusions. However, Dalldorf's [8] original observations have been confirmed by workers in many different parts of the country, indicating a wide distribution of this agent among human populations.[39] In contrast to the few examples of erratic rodent-adaptable strains of poliomyelitis virus, which have been mentioned before, it is obvious that we are dealing here with a very common agent which displays constant selective pathogenicity for suckling mice. It is included within the poliomyelitis group of viruses tentatively only because it shows striking differences when compared with the classic strains. On the other hand, within the relatively short time since its discovery it has been isolated with increasing frequency not only from the feces of patients with benign nonparalytic disease suspected to be abortive poliomyelitis but also from the feces of the typically paralyzed ones, occasionally even together with classic virus. According to a recent report, Coxsackie virus may even be recovered from the blood during the first week of illness or from the brain and spinal cord in fatal cases, though caution is needed in the interpretation of these results, because the agent may be disseminated in the laboratory.[40] The occurrence of specific antibodies and their rise in titer during the attack strongly suggests that the virus participates actively in the disease process. Finally, its geographic and seasonal distribution follow closely the distribution of

37. (*a*) Kling, C.; Levaditi, C., and Lépine, P.: Bull. Acad. de méd., Paris **102**:158, 1929. (*b*) Burnet, F. M., and Jackson, A. V.: Australian J. Exper. Biol. & M. Sc. **18**:361, 1940. (*c*) Sabin, A. B., and Ward, R.: Science **94**:113, 1941. (*d*) Melnick.[13]

38. Melnick, J. L.: Am. J. Hyg. **49**:8, 1949.

39. Curnen, E. C.; Shaw, E. W., and Melnick, J. L.: Disease Resembling Nonparalytic Poliomyelitis Associated with Virus Pathogenic for Infant Mice, J. A. M. A. **141**:894 (Nov. 26) 1949.

40. Howitt, B. F.: Proc. Soc. Exper. Biol. & Med. **73**:443, 1950.

Landsteiner's poliomyelitis virus. In other words, as far as is known at this time, Coxsackie virus is present as a disease-producing agent in cases which are clinically and epidemiologically indistinguishable from cases of classic poliomyelitis. More recently, however, Coxsackie virus has been encountered in outbreaks of diseases with widely divergent manifestations not suggestive of poliomyelitis.[41] The particle size of the virus as determined by ultrafiltration has been variously estimated, either as being 10 millimicrons or less [31] or between 15 and 23 millimicrons,[12] with a sedimentation constant of 135 to 155 Svedberg units. The virus produces flaccid paralysis in suckling rodents (mice, hamsters, guinea pigs, cotton rats), the characteristic feature of the experimental disease being a widespread, intense myositis in the affected limbs with no central nervous system lesions, or lesions of an indefinite type (encephalopathy) (fig. 2). The absence of typical brain and cord lesions may be due to a lack of response on the part of the infant central nervous system rather than to a lack of neurotropism of the virus itself, because the agent seems to multiply in the brain of the infected suckling mouse. It may prove significant that the various Coxsackie strains can be readily classified into two groups on the basis of the histological examinations of paralyzed suckling mice.[42] Group A includes

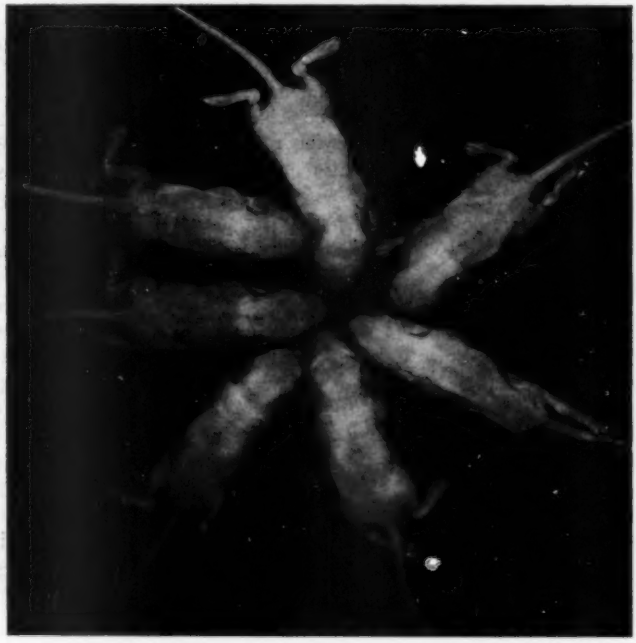

Fig. 2.—Flaccid paralysis induced in infant mice by intraperitoneal infection with Coxsackie virus isolated from human feces.

41. Kilbourne, E. D.: Federation Proc. **9**:581, 1950.

42. Dalldorf, G.: Federation Proc. **9**:569, 1950.

strains that induce lesions only of the skeletal muscles; group B strains also cause lesions in the brain and sometimes in the fat pads, heart and other organs. Pappenheimer and associates [43] have gone even further in proposing a separation of the different strains into four classes, defined according to their respective degrees of predilection for skeletal muscle, heart muscle, adipose tissue, abdominal viscera and central nervous system. Such divisions are valuable in that they emphasize the successive blending of non-nerve and nerve tissue tropisms for the several strains within this large group. It seems likely that conflicting opinions with regard to particle size and association with various clinical conditions may ultimately be resolved by realizing that Coxsackie viruses exist as a huge, basic group that includes member strains with distinctive yet overlapping etiologic significance. Attempts to produce clinical disease in monkeys by parenteral routes have failed. However, following the oral administration of Coxsackie virus, an infectious state could be established in cynomolgus monkeys and in chimpanzees but not in rhesus monkeys.[44] The type of response is quite similar to that observed in chimpanzees following the feeding of classic human poliomyelitis virus.

The precise relationship between murine Coxsackie virus and simian poliomyelitis virus is, of course, a matter of speculation. The possibility must be considered, however, that poliomyelitis virus may occur in two forms: (1) a primitive, nondifferentiated viscerotropic form, which possesses only sufficient pathogenicity to multiply on infantile muscle tissue, and (2) a highly specialized neurotropic form, which depends for its multiplication on environmental factors found only in human or simian nerve tissue. It is conceivable that these two forms represent the extreme end points in a process of selective tissue adaptation [45] for a virus which has given other circumstantial evidence of wide fluctuations in its degree of pathogenicity for man. The more conservative view, held by a majority of workers at present, would be that Coxsackie virus is a separate etiological agent and the cause of a new human disease.

II. PROBLEMS OF CLASSIFICATION

The multiplicity of murine strains which have been isolated (1) from cases of human poliomyelitis, (2) from presumable cases of human poliomyelitis and (3) from at present obscure sources with no immediate implication of the human disease is bewildering. As previously mentioned, all are morphologically similar with respect to particle size (except EMC and some strains of Coxsackie virus), allowing for the fact that measurements by ultrafiltration always give somewhat lower figures (10 to 15 millimicrons, average) than estimations from electron micrographs (20 to 35 millimicrons, average).[46] The various strains, however, show gradations in certain biologic properties which permit placing them in three separate, serologically distinct groups, i. e., the Lansing group, the Col.SK group and the Coxsackie group. Because all strains have been employed at times as experimental models of the agent responsible for the human disease, it is necessary to discuss in some detail their taxonomic position and the probable relationship to the agent or

43. Pappenheimer, A. M.; Daniels, J. B.; Cheever, F. S., and Weller, T. H.: J. Exper. Med. **92**:169, 1950.

44. Melnick, J. L., and Ledinko, N.: J. Immunol. **64**:101, 1950.

45. Jungeblut, C. W., and Steenberg, E.: Arch. Path. **49**:574, 1950.

46. Reagan, R. L.; Schenck, D. M., and Brueckner, A. L.: J. Infect. Dis. **86**:295, 1950.

agents which cause poliomyelitis in man, as the term is commonly understood on clinical, pathological and epidemiological grounds. This discussion must take into consideration the pathogenicity, the immunologic properties and the general biologic characteristics of the various strains.

PATHOGENIC PROPERTIES

Beginning with the problem of pathogenicity, it can be said that the murine strains of all three groups have varying degrees of pathogenicity for monkeys. This ranges from fully developed power to paralyze rhesus monkeys (Lansing group) or cynomolgus monkeys (Col.SK-MM-EMC-Mengo group) (figs. 3 to 6) to the capacity to infect the animal only inapparently, i. e., to induce fever, prolonged fecal excretion and specific antibody formation in cynomolgus monkeys (Coxsackie

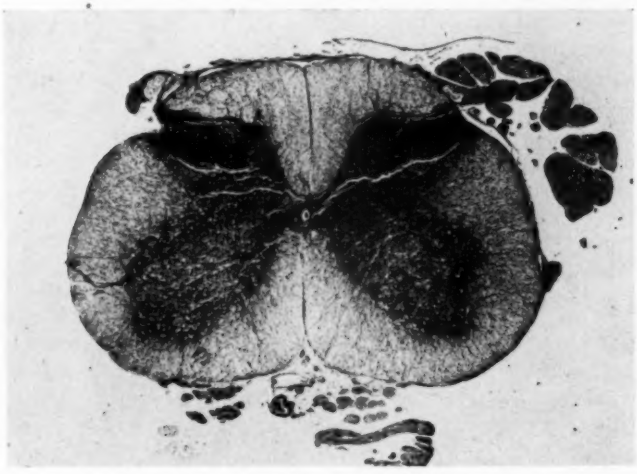

Fig. 3 (cynomolgus 1).—Section through lumbar cord. This monkey had paralysis of the left arm and leg six days after intracerebral injection of cavian Col.SK virus (eighty-first passage).

group). This basic property clearly differentiates these strains from the known strains within the Theiler (spontaneous mouse encephalomyelitis) group, none of which show any evidence of pathogenicity for either rhesus or cynomolgus monkeys.[30a] While monkey pathogenicity may deteriorate through prolonged mouse passages, this criterion is, perhaps, the most compelling argument in favor of accepting them as authentic murine strains of human virus.

Since the central nervous system has definite limitations in its ability to react to injury, interpretation of pathological data is beset with certain difficulties. Fortunately, however, the pathology of human and of experimental poliomyelitis gives, on the whole, a fairly consistent and characteristic picture which is determined by the type of lesions and the manner in which they are distributed throughout the spinal cord and certain areas of the brain. As Bodian states it, "As far as the

pathologist is concerned, all cases of poliomyelitis are 'encephalitic.' "[47] But, whereas other neurotropic viruses, especially those belonging to the epidemic encephalitis group (St. Louis, Japanese B, equine encephalomyelitis) or louping ill, may induce similar individual cell responses, the entire pattern presents features

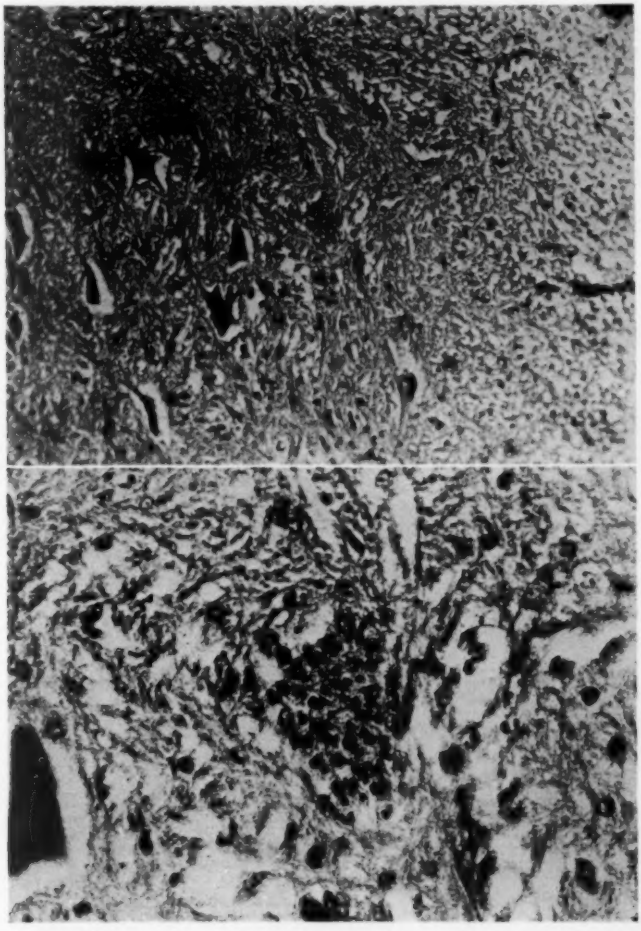

Fig. 4 (cynomolgus 1).—Upper, higher magnification of an area of an anterior horn shown in figure 3. Note the isolated destroyed nerve cell.
Lower, higher magnification of an area shown in upper figure, illustrating neuronophagia.

that make it unmistakably different and distinct from that found in poliomyelitic infection. Various experimental animals, however, may show dissimilar types of

47. Bodian, D., in Poliomyelitis: Papers and Discussions Presented at the First International Poliomyelitis Conference, 1948, Philadelphia, J. B. Lippincott Company, 1949.

response to poliomyelitis virus, even to the same strain, probably because of peculiarities of anatomic structure and metabolic function. Thus, monkeys when infected with Lansing (rhesus) or Col.SK (cynomolgus) virus show paralysis with typical acute poliomyelitic lesions in the cord, such as hemorrhage, perivascular

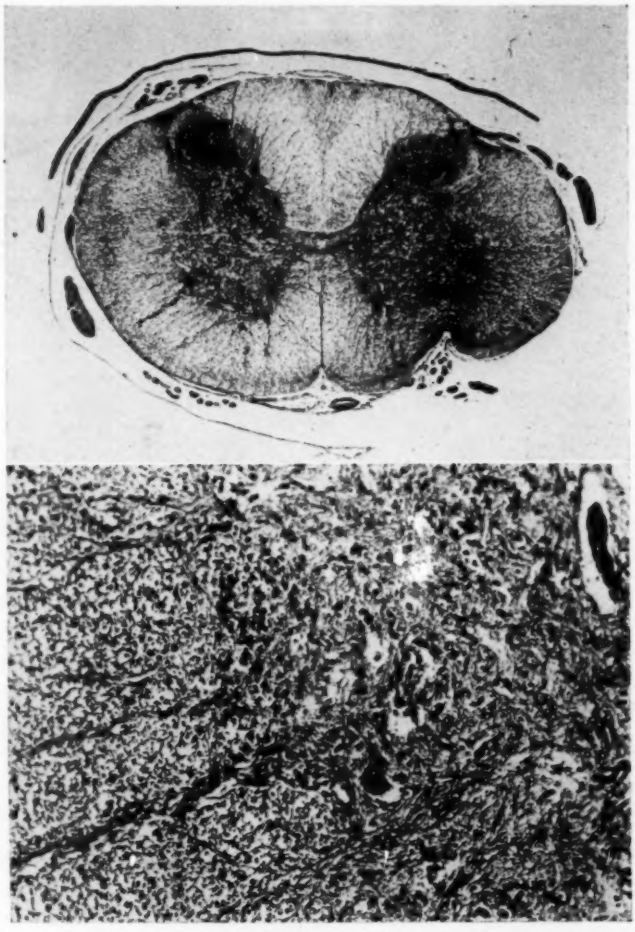

Fig. 5 (cynomolgus 5).—Upper, section through cervical cord. This monkey had paralysis of the right leg three days after intracerebral injection of Col.SK murine virus (four hundred twenty-second mouse passage).

Lower, higher magnification of an area of an anterior horn shown in upper figure. Note extensive destruction of nerve cells and perivascular cuffing.

cellular infiltration and necrosis of ganglion cells, often with well developed neuronophagia. Destruction of nerve cells and cuffing around blood vessels are frequently found also in the medulla and the pons. Cortical damage, as a rule, is limited to the

motor area and may or may not be present, depending on the portal of entry chosen and the virulence of the virus. In the cerebellum, in our experience, lesions may be present in the roof, but the cortical zone remains free, and the Purkinje cells are well preserved. Meningeal involvement is usually slight. The observed symptoms (flaccid paralysis, occasionally with transient encephalitic signs) correspond well

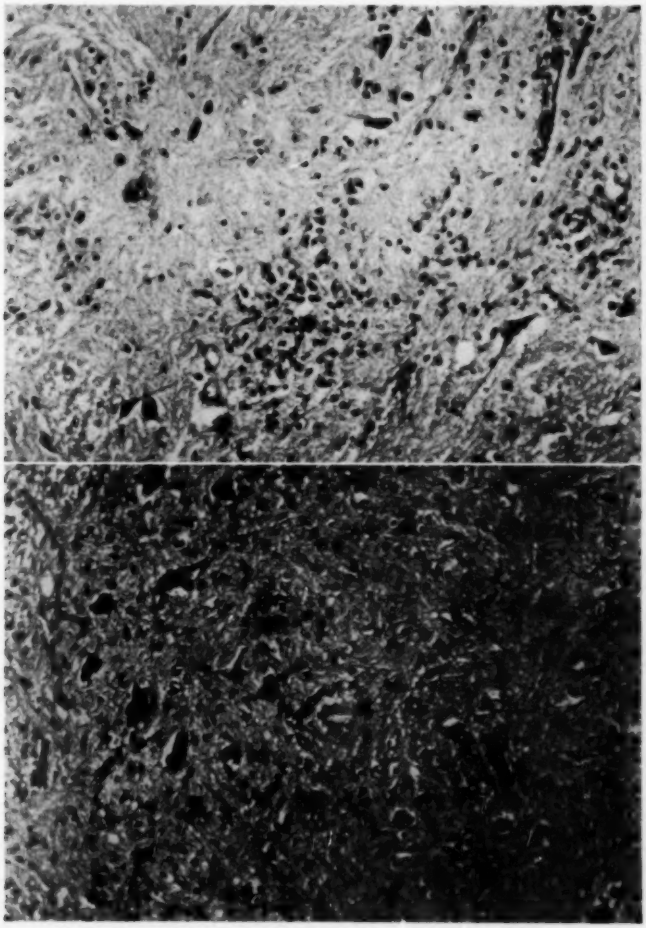

Fig. 6.—Upper, section through cervical cord of rhesus AS 60 showing nerve cell destruction and perivascular cuffing. This monkey had paralysis of the left arm, seven days after intracerebral injection of Col.SK cavian virus (second monkey passage, cynomolgus 7).

Lower, section through lumbar cord of cynomolgus 23 showing nerve cells in various stages of disintegration. This animal had generalized paralysis five days after intracerebral injection of EMC virus (second monkey passage, cynomolgus 17).

with the localization of the described lesions. These experimental findings are in good agreement with the pathology and symptomatology of the human disease.

A somewhat different picture, however, presents itself to the pathologist who studies the lesions induced in rodents by these same strains, especially the Lansing and the Col.SK virus, which have been closely examined.[48] In mice paralyzed by either strain the changes in the central nervous system are alike in that lesions of the anterior horn are fairly constant and of similar type, but the myelitis is overshadowed by an extensive involvement of the brain stem, the rhinencephalon, the cerebellum and the cerebral cortex. Hence, mice or cotton rats infected with these strains may show, in addition to flaccid paralysis, an encephalitic syndrome.[49] Yet this is not true of the histopathological changes induced by Col.SK virus in guinea pigs [50] or in hamsters.[51] Here, the anterior horns are the site of the severest damage, and lesions diminish progressively both in intensity and in frequency as one ascends the central nervous system. Cortical lesions, when present, are minor and usually associated with the inoculation tract; the olfactory bulbs show only isolated lesions, and the cerebellum is not involved at all. In other words, in the larger rodents, which exhibit much more resistance to the virus than the mouse, the distribution and histological detail of the lesions approach the pathology of simian and human poliomyelitis very nearly.

On reviewing this evidence one cannot escape the impression that the specific localization and the severity of the lesions that occur in the central nervous system in poliomyelitic infection depend, in a large measure, on factors connected with the resistance of the host and the invasiveness of the virus rather than on fundamentally divergent properties of individual strains.[52] This impression is also reflected by clinical studies, which show a wide diffusion of spinal paralytic forms (poliomyelitis), bulbar forms (polioencephalomyelitis), encephalitic forms (polioencephalitis) and mixed forms, often in the same family.[53] In a recent review of the problem the British Ministry of Health [54] made a proposal "to supersede the regulation requiring the notification of polioencephalitis as a separate clinical entity and to revert to the single generic term 'poliomyelitis' (syn. polio-encephalomyelitis)."

Pathological involvement of the peripheral nervous system—that is, peripheral nerve, motor end plate and contiguous muscle fibers—in poliomyelitis is still under discussion. While there can be little disagreement that functional disturbances at the myoneural junction do occur in the paralyzed muscle, it is debatable whether these changes are primary or secondary. So far as structural damage to nerve endings

48. Lillie, R. D., and Armstrong, C.: Pub. Health Rep. **55**:115, 1940; **55**:718, 1940. Wolf, A.: J. Exper. Med. **76**:53, 1942.

49. Vieuchange, J.: Ann. Inst. Pasteur **73**:1133, 1947. Vieuchange, J., and Descola, P.: Ibid. **73**:1135 and 1137, 1947. Bodian, D.; Morgan, I. M., and Schwerdt, C. E.: Am. J. Hyg. **51**:126, 1950.

50. (*a*) Jungeblut, C. W.; Feiner, R. R., and Sanders, M.: J. Exper. Med. **76**:31, 1942. (*b*) Schultz, E. W., and White, S. C.: Proc. Soc. Exper. Biol. & Med. **68**:266, 1948.

51. Dalldorf, G., and Whitney, E.: Proc. Soc. Exper. Biol. & Med. **59**:150, 1945.

52. Janings, G. H.; Hamilton-Petterson, J. L., and MacCallum, F. O.: Brit. M. J. **2**:210, 1949.

53. Fanconi, G.; Zellweger, H., and Botsztejn, A.: Die Poliomyelitis und ihre Grenzgebiete, Basel, Benno Schwabe & Co., 1945. McQuarrie, I., in Poliomyelitis: Papers and Discussions Presented at the First International Poliomyelitis Conference, 1948, Philadelphia, J. B. Lippincott Company, 1949. Baker, A. B., in Poliomyelitis: Paper and Discussions Presented at the First International Poliomyelitis Conference, 1948, Philadelphia, J. B. Lippincott Company, 1949. Pohl, J. F.: Arch. Pediat. **66**:537, 1949.

54. Murray, L. H.: Brit. M. J. **2**:1028, 1947.

or to contractile muscle elements is concerned, peripheral lesions, located at the motor end plate [55] or within the muscle fiber,[56] have been described from time to time in human cases (acute and chronic) of the disease. Similar muscular lesions, consisting of necrosis and fragmentation of fibers with focal inflammatory response, occur in the heart muscle in fatal cases.[57] Again, the proper interpretation of these observations is open to some doubt. However, the fact that the active virus may be recovered from the paralyzed skeletal muscle (by biopsy) during the acute stage [58] as well as from the heart muscle at autopsy [59] carries weight in favor of regarding the pathological manifestations in the muscle as being due to primary activity of the virus. This conclusion is supported, finally, by the demonstration that human poliomyelitis virus can be successfully grown on extraneural tissue (intestine, muscle, skin, testis and others) in suitable human tissue culture mediums. If one searches for corresponding extraneural lesions in experimentally infected animals, conflicting results are obtained, especially with the various murine stains. Thus, infection of monkeys (cynomolgus) and rodents (mouse, guinea pig, hamster) with the Col.SK group of viruses is regularly accompanied with the occurrence of myositic (and central) lesions in the skeletal or heart muscle.[60] These lesions are indistinguishable from those produced by the Coxsackie group of viruses. By contrast, muscular lesions are conspicuously absent in monkeys or rodents paralyzed after infection with the Lansing group of viruses. Inasmuch as human poliomyelitis virus has also given some direct evidence of possessing both myotropic and neurotropic qualities, it would seem that the viscerotropic-neurotropic strains of the Col.SK group reproduce more faithfully in experimental animals the complex pathognomonic syndrome of the human disease than the neurotropically fixed Lansing group of strains.[29b] Perhaps so-called poliomyelitis virus exists in the form of an incomplete, latent, pantropic precursor that develops into a complete, paralyzing, neurotropic agent only on accidental or experimental contact with certain neural tissue components.

IMMUNOLOGIC PROPERTIES

Immunologic relationships with human poliomyelitis may be demonstrated by means of in vivo or in vitro methods. The former depend on the fact that monkeys paralyzed by known poliomyelitis virus will resist challenge with the test virus or that the test virus cannot be successfully recovered from the reinfected animal. Such relationships have been well established for the Lansing virus and are also obtainable with Col.SK virus.[61] However, they may partly depend on nonspecific protection by interference rather than on specific immunity mechanisms. In contrast to cross infection experiments, in vitro neutralization of virus by antiserum measures

55. Sanz Ibanez, J. trab. Inst. Cajal de inv. biol. **37**:259, 1945.

56. Denst, J., and Neuberger, K. T.: Am. J. Path. **26**:863, 1950.

57. (*a*) Saphir, O., and Wile, S. A.: Am. J. M. Sc. **203**:781, 1942. (*b*) Dolgopol, V. B., and Cragan, M. D.: Arch. Path. **46**:202, 1948. (*c*) Ludden, T. E., and Edwards, J. E.: Am. J. Path. **25**:357, 1949.

58. Jungeblut, C. W., and Stevens, M. A.: Am. J. Clin. Path. **20**:701, 1950.

59. (*a*) Jungeblut, C. W.: Proceedings of the Third International European Poliomyelitis Conference, Amsterdam, 1950. Jungeblut, C. W., and Edwards, J. E.: Am. J. Clin. Path., to be published.

60. (*a*) Rustigian, R., and Pappenheimer, A. M.: J. Exper. Med. **89**:69, 1949. (*b*) Jungeblut and Steenberg.[45] Jungeblut, C. W.: Unpublished data.

61. Jungeblut, and Sanders.[4] Jungeblut, C. W.: Unpublished data.

a finer degree of antigenic specificity which may vary considerably from strain to strain. Individual serologic strain properties, therefore, contribute little or nothing to the heuristic value of the over-all identification scheme. Thus, the predominating group of monkey-pathogenic strains, which cannot be adapted to rodents, exist in at least three distinct serologic types,[62] and more will probably be added, as new, fresh human strains are being isolated and studied. The serologic relationships found for the neurotropic murine strains within the first group appear to be simple, since cross neutralization is readily obtained between Lansing, MEF, Y-SK and even WW virus.[63] A similar homogeneity is found with the neurotropic-viscerotropic murine strains comprising the second group (Col.SK-MM-EMC-Mengo). On the other hand, the otherwise so compact Coxsackie group apparently is split into an array of strains with multiple antigenic complexion.[64] Antigenic variation on such a scale is precisely what one might expect from a primitive, undifferentiated agent. The three groups seem to be serologically quite independent, even though unilateral cross reactions may be demonstrated between Col.SK and Y-SK virus when highly potent antiserums are used. These reactions, however, are strictly limited to Y-SK antiserum inactivation of Col.SK virus, whereas Col.SK antiserum has no effect whatsoever on Y-SK virus.[65] The results indicate that the neurotropic strains are highly specialized agents which require homologous antibody for their inactivation.

Notwithstanding their serologic variability, however, all viruses in the three basic groups possess a definite antigen component which can be neutralized by polioconvalescent human serum, a fact which adds circumstantial evidence to their imputed human pathogenicity. This phenomenon, presumably specific, is easily realized with viruses of the Lansing type, running up to between 50 and 75 per cent neutralization in unselected series of human polioconvalescent serums. It is also well established for the Coxsackie virus but is complicated here by the existence of sharply differing serologic types. Neutralization of the Col.SK-MM-EMC group of viruses by human convalescent serum is definitely more limited. Thus, Smadel and Warren [66] reported significant antibody levels against EMC virus in 17 of 44 serums examined. These serums were from military patients hospitalized in Manila (1945-1946) with a clinical diagnosis of "three day fever" (aseptic meningitis? abortive poliomyelitis?). In a larger series of over 100 serums obtained from patients whose illness was diagnosed as paralytic or abortive poliomyelitis during the epidemic of 1949 in Greater New York, about 20 per cent gave positive neutralization of Col.SK virus, whereas approximately 55 per cent neutralized Y-SK virus.[67] Since the frequency of such neutralization is conventionally regarded as a numerical indicator of the epidemic or endemic distribution of a given strain, it follows that Col.SK virus is not a common infecting strain in human populations. The significance of this conclusion is obvious, but more data are needed, possibly with the aid of hemagglutination inhibition tests,[68] in order to interpret properly its

62. Bodian, D.; Morgan, I. M., and Howe, H. A.: Am. J. Hyg. **49**:234, 1949. Kessel, J. F., and Pait, C. F.: Proc. Soc. Exper. Biol. & Med. **70**:315, 1949.

63. Levaditi, C., and Vaisman, A.: Compt. rend. Soc. de biol. **143**:360, 1949.

64. Sickles, G. M., and Dalldorf, G.: Proc. Soc. Exper. Biol. & Med. **72**:30, 1949.

65. Jungeblut, C. W.: To be published.

66. Smadel, J. E., and Warren, J.: J. Clin. Invest. **26**:1197, 1947.

67. Jungeblut, C. W.: Arch. Pediat. **67**:519, 1950.

68. (*a*) Gard, S., and Heller, L.: Proc. Soc. Exper. Biol. & Med. **76**:68, 1951. (*b*) Jungeblut, C. W., and Horvath, B.: Federation Proc. **10**:411, 1951.

full epidemiological implication. Yet, for all practical purposes, demonstration of some degree of characteristic monkey pathogenicity and neutralization by certain samples of human convalescent serum remain as the only practical procedures for the tentative identification of a given rare strain isolated from material presumably containing human poliomyelitis virus.

BIOLOGIC PROPERTIES

A number of other features must be discussed which separate the murine strains of the Lansing group from those cataloged as the Col.SK-MM-EMC-Mengo group. Foremost among these is the high virulence for rodents (mice, guinea pigs, cotton rats, hamsters), which is shared by all members of this group, as contrasted with the low grade infectivity of the Lansing type. Peripheral infection with the Lansing strain, for instance, does not succeed in rodents except in suckling cotton rats under 12 days of age.[69] Curiously enough, suckling mice are relatively refractory to any mode of infection with virus of the Lansing type,[70] whereas it is precisely this animal which is so selectively susceptible to infection with Coxsackie virus. Another differential point concerns the extent to which virus cultivation is possible in embryonated hen's eggs or in surviving tissue culture mediums. Col.SK, MM and Mengo murine virus have been successfully propagated in the chick embryo [71] and in embryonic mouse tissue culture [26] on neural as well as on nonneural tissues; however, the less virulent cavian Col.SK virus will not multiply in the egg.[71b] The Lansing virus cannot be grown by either of these methods, but propagation of this virus has been achieved in human embryonic as well as in postembryonic tissue culture medium. Again, multiplication occurs with nervous as well as with non-nervous tissue.[72] This is also true for other simian strains of human virus as well as for MM virus.[73] Coxsackie virus, finally, has been grown in the embryonated egg [74] and has been cultivated in mouse culture medium.[75] A third differential characteristic has evolved from hemagglutination studies. Thus, the Col.SK-MM-EMC-Mengo group of viruses will agglutinate sheep red cells, a phenomenon which is not readily demonstrated with the Lansing virus.[76] The reasons for this failure are not clear and may conceivably be found in differences in quantity rather than quality of virus.[25b] Moreover, fresh human strains of poliomyelitis virus have as yet not been studied

69. Pinto, M. E.: Am. J. Hyg. **48**:361, 1948.

70. Sabin, A. B.: Proc. Soc. Exper. Biol. & Med. **73**:394, 1950.

71. (*a*) Schultz, E. W., and Enright, J. B.: Proc. Soc. Exper. Biol. & Med. **63**:8, 1946. (*b*) Enright, J. B., and Schultz, E. W.: Ibid. **66**:541, 1947. (*c*) Powell, H. M., and Jamieson, W. A.: J. Infect. Dis **83**:238, 1948. (*d*) Dick, G. W. B.: Proc. Soc. Exper. Biol. & Med. **73**:77, 1950.

72. (*a*) Enders, J. F.; Weller, T. H., and Robbins, F. C.: Science **109**:85, 1949. (*b*) Weller, T. H.; Robbins, F. C., and Enders, J. F.: Proc. Soc. Exper. Biol. & Med. **72**:153, 1949. (*c*) Milzer, A.; Levinson, S. O.; Vanderboom, K., and Adelman, P.: Ibid. **74**:136, 1950. (*d*) Smith, W. M.; Chambers, V. C., and Evans, C. A.: Northwest Med. **49**:368, 1950.

73. Chambers, V. C.; Smith, W. M., and Evans, C. H.: Proc. Soc. Exper. Biol. & Med. **76**:213, 1951.

74. Slater, E. A., and Syverton, J. T., Proc. Soc. Exper. Biol. & Med. **74**:509, 1950.

75. Huebner, R. J.; Ransom, S. E., and Beeman, E. A.: Pub. Health Rep. **65**:803, 1950.

76. Hallauer, C., in Proceedings of the Fourth International Congress for Microbiology, 1947, Copenhagen, Rosenkilde & Bagger, 1949. Verlinde, J. D., and de Baan, P.: Ann. Inst. Pasteur **77**:632, 1949. Olitsky, P. K., and Yager, R. H.: Proc. Soc. Exper. Biol. & Med. **71**:719, 1949. Horvath, B., and Jungeblut, C. W.: Federation Proc. **10**:359, 1951.

for possible hemagglutination. All in all, the whimsical character of this reaction is demonstrated by the fact that Theiler's GDVII strain will agglutinate human O red cells, whereas Theiler's FA strain fails to do so.[77] Both strains are biologically and immunologically closely related; in fact, GDVII hemagglutination can be specifically inhibited by GDVII immune serum as well as by FA immune serum.

The above mentioned differences in biologic properties provide a basis for formally separating the Lansing group from the Col.SK-MM-EMC-Mengo group and from the Coxsackie group of viruses. Few of these differences, however, are due to mutually exclusive properties, and most appear to be more quantitative than qualitative in character. It seems not unreasonable to assume, for instance, that potency, peripheral infectivity, ability to grow on or become attached to certain cells, and power to invade the blood stream reflect various degrees of inherent virulence or tropism of the infectious agent for the substrate of its host. This tropism may be unevenly developed with different strains and may undergo gradual and successive evolution within a large family of simian and rodent poliomyelitis viruses. Experimental evidence for this concept may be found in the characteristic biphasic (Lansing) or monophasic (Col.SK, Coxsackie) distribution of the incubation periods in mice, which suggests basic differences in homogeneity among various strains with respect to their content of monkey-pathogenic and rodent-pathogenic components.[78]

Little is known as to whether variation of poliomyelitis virus capable of transforming one type of strain into another may occur, and the available evidence is confusing. While a number of workers have stressed a remarkable stability of type, examples are not wanting to suggest that such variation may be possible, at least in principle.[79] Unfortunately, there is no certainty that "high" strains—believed to have originated through variation—are actually derived from "low" strains, because experiences of this sort are isolated and the results not repeatable at will. Some of such "high" strains may well be Theiler strains, with uncommon serologic properties, picked up inadvertently [80]; others, which are serologically related to Col.SK virus, may have come about through accidental contamination with a member strain of the Col.SK-MM-EMC-Mengo group of viruses carried at the same time in the laboratory. The chances for this to occur are rather remote, though, inasmuch as natural spread of Col.SK and MM virus has not been observed except under forced conditions of contact that do not apply to routine laboratory experimentation.[81] An intriguing observation which points to the inherent variability of SK virus, at least, was made in this laboratory.[82] When Y-SK virus is injected

77. Labelle, O., and Horsfall, F. L.: Proc. Soc. Exper. Biol. & Med. **71**:713, 1949.

78. (*a*) Young, L. E., and Merrell, M.: Am. J. Hyg. **37**:80, 1943. (*b*) Salk, J. F., and Bennett, B. L.: J. Immunol. **66**:283, 1951.

79. Jungeblut, C. W.: Am. J. Pub. Health **33**:1227, 1943; Science **99**:434, 1944. Gallia, F.: Bol. Inst. invest. vet. **3**:473, 1947. Jungeblut, C. W., in Proceedings of the Fourth International Congress for Microbiology, 1947, Copenhagen, Rosenkilde & Bagger, 1949. Milzer, A., and Bryd, C. L.: Science **105**:70, 1947. Sanders, F. K.: Federation Proc. **7**:309, 1948. Lawson, R. B., and Melnick, J. L.: J. Infect. Dis. **79**:201, 1947. Kling, C.; Levaditi, C., and Vaisman, A.: Compt. rend. Soc. de biol. **142**:1348, 1948. Behrend, R. C., and Schultz, H. W.: Klin. Wchnschr., to be published. Enright and Schultz.[71b]

80. Melnick, J. L., and Riordan, J. T.: J. Immunol. **57**:331, 1947.

81. Annual Report, Division of Laboratories and Research, New York State Department of Health, Albany, New York, 1946. Schultz and White.[50b]

82. Jungeblut, C. W.: Unpublished data.

into suckling mice, the virus, as a rule, fails to produce paralysis and cannot be carried in passage. On one occasion, however, paralysis developed in some suckling mice following intracerebral injection of Y-SK virus. The agent could be serially transmitted in suckling mice over five consecutive passages by either intracerebral or intraperitoneal injection of high dilutions of brain (10^{-6} to 10^{-8}), with the production

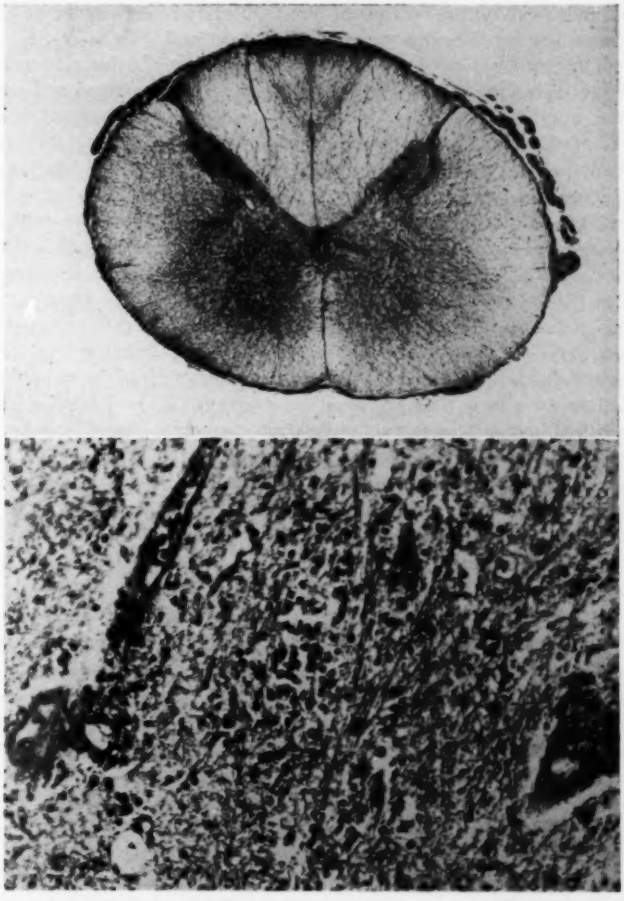

Fig. 7 (rhesus AR 97).—Upper, section through lumbar cord. The monkey had complete paralysis of the left leg six days after intracerebral injection of Y-SK mouse virus (third suckling mouse passage).

Lower, higher magnification of an area of an anterior horn, shown in upper figure, illustrating perivascular infiltration and nerve cell destruction.

of flaccid paralysis. The virus also had the same potency in young adult mice. When the virus was tested for monkey pathogenicity, typical flaccid paralysis—with extensive poliomyelitic lesions of the cord—was regularly obtained in rhesus

monkeys with intracerebral injection of virus harvested from successive suckling mouse passages (I to V) (fig. 7). Monkey convalescent serum had antibodies for both Y-SK and Col.SK virus, but the suckling mouse virus, in remote serial passage, was significantly inactivated only by Col.SK antiserum. Control experiments were set up which demonstrated conclusively (1) that Col.SK virus alone, while paralyzing suckling mice, was not rhesus pathogenic and (2) that prepared mixtures of Col.SK and Y-SK virus, because of effective interference, cannot be carried in suckling mice beyond the first passage without a total loss of Y-SK virus, as determined by serologic tests and the lack of pathogenicity for rhesus monkeys. An agent was therefore obtained in this instance which combined and maintained some of the properties of Col.SK and of Y-SK virus, apparently fused in one stem virus.[83] It would seem that the last word has not yet been said on the problem of variations of poliomyelitis virus and final judgment should be suspended until additional information is at hand.

Finally, it is well to remember that certain neurotropic viruses—morphologically and with respect to their host spectrum quite distinct from poliomyelitis virus—may imitate the clinical picture of poliomyelitis in man and even possess features suggesting some general relationship with the poliomyelitis group. We are referring here to the virus of the so-called Australian X disease, the Japanese B encephalomyelitis virus and possibly the virus of louping ill. In 1918 Breinl [84] studied an outbreak of a "mysterious disease" in Australia which was characterized by a predominance of encephalitic symptoms over paralytic involvement. In two fatal cases of this polioencephalitis nerve tissue was transferred to cercopithecus monkeys. The resulting experimental disease was, in symptoms and in pathology, identical with that produced by inoculation of material from undoubted cases of poliomyelitis. The author stated: "The conclusion is justified that the cases of 'mysterious disease' occurring in Queensland and New South Wales were caused by the same virus as acute poliomyelitis." Unfortunately, the virus was lost and is therefore not now available for comparative study. The agent has subsequently been regarded either as Japanese B encephalomyelitis virus or as the virus of louping ill, largely because of the presence of cerebellar lesions and its pathogenicity for sheep. In the case of Japanese B encephalitis virus, the most comprehensive antigenically within the encephalitis group, the following features command attention: (1) Pathological changes in the spinal cord, resembling those produced by poliomyelitis virus, have been described in some fatal cases [85]; (2) virucidal antibodies have been found in

83. An analogous phenomenon has recently been described by W. M. Hammon (Am. J. Trop. Med. **28**:515, 1948) for the encephalitis group of viruses. From the standpoint of epidemiology, pathology and clinical symptoms as observed in man Western equine encephalitis virus and St. Louis encephalitis virus are remarkably similar. The two viruses are antigenically distinct, but antibodies against both viruses are frequently found together in human and animal serums. The number of these apparent "double infections" is too high to be considered as due to chance alone. Out of mites collected from a wild bird's nest there was isolated an agent, or a mixture of agents, which had the characteristics of both Western equine virus and St. Louis virus. The author postulates the existence of a stem virus and suggests that from this ancestral stem, as it passes through mammalian or avian hosts, one or another mutant may assume dominance.

84. Breinl, A.: M. J. Australia **1**:209, 1918.

85. Zimmerman, H. M.: Am. J. Path. **22**:965, 1946. Sabin, A. B.; Schlesinger, R. W.; Ginder, D. R., and Matumoto, M.: Am. J. Hyg. **46**:356, 1947.

normal human and animal serums in areas of the world in which Japanese B encephalitis has not occurred.[86] It is not clear whether the so-called Japanese B virus may have subgroups with properties resembling those of viruses within the poliomyelitis group or whether the diagnosis of Japanese B encephalitis has at times been confused with that of polioencephalitis. Further work is necessary to clarify this point.

A SYSTEM OF CLASSIFICATION

The nomenclature and the classification of poliomyelitis are still far from satisfactory because the well known clinical (infantile paralysis) and anatomic (acute anterior poliomyelitis) terms do not adequately describe the disease in its manifold appearances. Realizing the need for an etiological approach to the problem, Wickman [87] suggested "that all forms of the disease arising from the same virus as acute poliomyelitis should be grouped under the one term—Heine-Medin's disease," after the two pioneer workers in the field. Unfortunately, the problem of classifying the etiological agent is inextricably tied up with the even more formidable problem of classifying the disease. For, as has previously been said, few infectious diseases seem more complex in their epidemiological, clinical and pathological manifestations than what is currently called poliomyelitis. However, a great deal more is known about the virus since Wickman's early suggestion. Therefore, an attempt seems justified at this time to devise a semiformal system of classification of poliomyelitis virus, the principal objective of which is to reconcile the established, older observations with the multitude of newer facts. To be of constructive value, such a system ought to stress similarities instead of dissimilarities, giving preference to fundamental principles rather than singling out isolated features. Above all, it should not be rigid but should be sufficiently flexible to accommodate expansion, bound to occur in a rapidly progressing field of research. While liberal in its concept, it must, nevertheless, recognize certain critical lines of demarcation which are generally accepted as valid criteria in systematic nomenclature. The proposed system of classification, therefore, differs from previous taxonomic efforts deliberately in the sense that it abandons homogeneity of the virus and introduces—as valid governing principles—phenomena of inherent variability of the etiological agent with respect to tissue reactivity and antigenic composition.

For the purpose of this discussion, then, poliomyelitis virus is defined as a viral agent, trypsin-resistant and ether-resistant, of approximately 8 to 15 millimicron size. The virus is capable of setting up a febrile infectious state in the intestinal tract, accompanied by fecal excretion, or, of inducing inflammatory and necrotizing lesions in the motor neuron. These lesions may appear at any point along the route of the motor neuron, i. e., the motor cortex, the brain stem, the anterior horn cell, the peripheral nerve, the myoneural junction and the peripheral muscle fiber. Different strains vary in their ability to produce central and/or peripheral lesions (in conjunction with similar variations in the clinical picture and the pathology of the human disease). Individual strains show, furthermore, differences in the extent of their pathogenicity for monkeys (various

86. Sabin, A. B.: St. Louis and Japanese B Types of Epidemic Encephalitis: Developments of Noninfective Vaccines; Report of Basic Data, J. A. M. A. **122**:477 (June 19) 1943. Koprowski, H.: J. Immunol. **54**:387, 1946.

87. Wickman, O. I.: Acute Poliomyelitis, translated by W. J. M. A. Maloney, Nervous and Mental Disease Monograph 16, New York, Nervous and Mental Disease Monographs, 1913.

species) and rodents. Existing strains may also occur in form of antigenically differing subtypes. Because of the varying and perhaps shifting properties associated with the virus with respect to tissue tropism and host spectrum, it is proposed to recognize a "family of poliomyelitis viruses" which includes groups of strains causing natural disease in man or in animals and possessing one or the other, or a combination, of the fundamental properties indicated above. The

Human Poliomyelitis Virus

Criteria of Identification	Poliomyelitis Virus, Simian Group	Para-Poliomyelitis Virus: Simian-Murine Group A Lansing Type	Para-Poliomyelitis Virus: Simian-Murine Group B Col.SK Type	Pseudo-Poliomyelitis Virus, Murine Group Coxsackie Type
Physical properties				
Heat inactivation	52.5 C.	50-55 C.	56.5 C.	60 C.
Ether and trypsin resistance	Resistant	Resistant	Resistant	Resistant
Size				
Ultrafiltration	8-17 millimicrons	8-17 millimicrons	9-14 millimicrons	6-9 millimicrons; 15-23 millimicrons
Electron microscopy	20-50 millimicrons	12-34 millimicrons	20-35 millimicrons	?
Sedimentation constant	125-130 Svedberg units	125	130 (Col.SK); 153 (EMC)	135-155
Biologic properties	Fecal excretion	Fecal excretion ?	Fecal excretion	Fecal excretion
Experimental disease				
Primates:				
Chimpanzee	+	Subclinical infection (by feeding virus)	Not done	Subclinical infection (by feeding virus)
Rhesus monkeys	+	+	Subclinical infection §	Negative
Macacus cynomolgus	+, ++, +++ †	+	+, ++, +++	Subclinical infection (by feeding virus)
Cercopithecus	+	+	+, ++	Not done
Cebus capucina	+ (fresh strains)	Negative	+ ?	Not done
Rodents:				
Infant mouse	Negative	± to +	+, +++	+, ++? +++
Infant cotton rat	Negative	+	+	+, +++
Infant hamster	Negative	+	+	+, +++
Infant guinea pig	Negative	Not done	+, +++	+, +++
Adult mouse	Negative	+	+ (++) +++	Negative
Adult cotton rat	Negative	+	+	Negative
Adult hamster	Negative	+	+	Negative
Adult guinea pig	Negative	Subclinical infection ‡	Clinical or subclinical infection	
Cultivation	Human tissue	Human tissue	Mouse tissue; chick embryo; human tissue	Mouse tissue; chick embryo
Immunologic characteristics				
Neutralization by convalescent human serums	Percentage of serums giving positive neutralization depending on serologic types	55-75% positive serums in unselected series	20% positive serums in unselected series	Percentage of positive serums depending on serologic types
Relationship to homologous human virus	Established	Established	Not clearly established but reasonably assumed	Established

The viruses listed under poliomyelitis virus and para-poliomyelitis virus are nonpathogenic for rabbits, dogs, cats, sheep and swine.

† See footnote [30b]

‡ Jungeblut: Unpublished data.

§ Paralytic infection with F virus.

\+ = Paralysis with central nervous system lesions.

++ = Cardiac lesions.

+++ = Skeletal muscle lesions.

essential properties of the strains listed under the "human group" are brought together in the table. The over-all classification scheme is as follows:

POLIOMYELITIS FAMILY

I. HUMAN VIRUS (neutralizable with human serum and potentially pathogenic for man)

A. *Poliomyelitis virus.* Simian strains (pathogenic for primates only), essentially neurotropic, especially after intracerebral passage fixation: (1) Aycock; (2) Brunhilde; (3) Leon; (4) other serologic types

B. *Para-poliomyelitis virus.* Simian-murine strains (pathogenic for primates and for rodents, either by experimental adaptation or by primary isolation).
   1. Low strains. Strains essentially neurotropic. Lansing, Y-SK, MEF, WW.
   2. High strains. Strains neurotropic and viscerotropic. Col.SK/MM group; EMC/Mengo group.

C. *Pseudo-poliomyelitis virus.* Murine strains (pathogenicity for infant rodents and limited pathogenicity for primates), Coxsackie ("C") virus.
   1. Strains only viscerotropic (with serologic subtypes).
   2. Strains viscerotropic and neurotropic (with serologic subtypes).

II. Mouse Virus (neutralizable with mouse serum and pathogenic for rodents)
   1. Neurotropic strains. O. T. Theiler.
   2. Neurotropic and viscerotropic strains. GDVII; FA.

III. Swine Virus (neutralizable with swine serum and pathogenic for swine) Teschen disease.

The above system follows, in principle, a scheme which was originally advanced by Gard.[88] It is also in general agreement with the classification adopted tentatively in Bergey's Manual published by the Society of American Bacteriologists,[89] as well as with recommendations voted by a conference on Nomenclature held in conjunction with the Fourth International Congress of Microbiology. The same underlying thoughts have finally been expressed in the discussions by Lépine (1949), by Mollaret (1950),[90] by Laruelle (1950) and by Verlinde (1950) at the occasion of the Second and Third International European Poliomyelitis Conferences. Some comment, however, is necessary concerning agreement or lack of agreement with a provisional classification proposed, in 1948, by a Committee on Nomenclature of the National Foundation for Infantile Paralysis.[91] The diagnostic criteria drafted by this committee for accepting a given virus as poliomyelitis virus were, in essence, as follows: "1. The term 'poliomyelitis virus' should be used to designate strains of the agent originally described as the cause of poliomyelitis in man, regardless of the source from which it may be recovered in nature. 2. The virus is identified by the typical experimental disease it induces in monkeys, by the character and the distribution of the microscopic lesions it produces in the central nervous system, by its host range, by its immunologic relationships and by its physicochemical properties. 3. Any virus which is immunologically distinct from any previously established poliomyelitis virus but which possesses the above-mentioned diagnostic properties must, nevertheless, be considered as a poliomyelitis virus." Since—subsequent to the publication of these recommendations—it was shown that the Col.SK group of viruses produces clinically and pathologically typical poliomyelitis in cynomolgus monkeys, it would appear that the principal diagnostic criteria have been met. This applies particularly if one assumes that these agents represent immunologically distinct virus types, originally of rodent descent, though potentially pathogenic for man, which may well stand between the native human and the rodent group. Hence, the rationale of Verlinde's proposal to recognize Col.SK virus as a fourth immunologic type of human poliomyelitis virus.[30c] There remains, however, a difference in opinion on the advisability of (1) setting up a phylogenetic classification including all viruses which

88. Gard, S.: Acta med. scandinav. 1943, supp. 143, p. 1.

89. Bergey, D. H.: Manual of Determinative Bacteriology, edited by R. S. Breed; E. G. D. Murray, and A. P. Hitchens, ed. 6, Baltimore, Williams & Wilkins Company, 1948.

90. Mollaret, P.: Presse méd. **58**:1096, 1205, 1223, 1255, 1950.

91. Armstrong, C., and others: A Proposed Provisional Classification of Poliomyelitis Virus, Science **108**:701, 1948.

naturally produce acute anterior poliomyelitis in man, in rodents and in swine, and (*2*) recognizing a group of human viruses, capable of producing poliomyelitis or poliomyelitis-like disease in man, which are selectively pathogenic for infant rodents, induce no characteristic central nervous system lesions and possess but limited pathogenicity for monkeys. Obviously, it is difficult to choose between two schemes which are constructed on fundamentally different biologic approaches. However, the unitarian viewpoint not only has the advantage of expressing gradations in viral activity which seems to occur in fluid sequence. It also obviates the dilemma of having to provide constantly for new viral entities and new diseases as strains with aberrant properties are being isolated.

CONCLUSIONS

As information concerning the mechanism of infection in human and experimental poliomyelitis accumulates, it seems advisable to review critically the properties of the virus itself. A survey of the data which have been made available through a variety of different approaches suggests that the agent which causes Heine-Medin disease is more complex than has hitherto been thought. In fact, it would appear that there exists a group of poliomyelitis or poliomyelitis-like disease entities which are caused by a corresponding group of distinct yet somehow related viral agents. It may therefore be of advantage to abandon the conventional definition of a single, homogeneous poliomyelitis virus and to recognize instead a family of poliomyelitis viruses which includes strains, morphologically similar, but dissimilar with respect to their antigenic structure and tissue tropism. This proposal has in its favor a number of recently discovered experimental facts that cannot be overlooked or quickly modified. Against it are the arguments of tradition, which many may not be willing to sacrifice at this time. The proposed classification scheme is therefore offered only as a working hypothesis and point of departure for further efforts to bring about eventually a comprehensive nomenclature for "poliomyelitis virus."

COEXISTENT GASTRODUODENAL AND CEREBRAL LESIONS IN INFANCY AND CHILDHOOD

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IN A SERIES of 251 consecutive autopsies on infants and children I observed one case of multiple acute ulcers of the stomach, two cases of gastromalacia, and seven of acute duodenal ulcer; in all these cases the lesion was associated with cerebral damage. Two additional cases of peptic ulcer were not included in this report since no primary cerebral lesions were present; a large subacute duodenal ulcer was found in an infant 10 weeks old and a small acute duodenal ulcer in a child of 3 months. Each had a history of long-standing diarrhea and inanition.

The literature on peptic ulcer in infancy and childhood has been reviewed by several authors.[1] In 1944 Hutchins [2] could find only 243 cases in the world literature. The relative infrequency with which these lesions have been observed in the past is indicated by the published reports. In 1913 Holt [3] found that among 1,800 autopsies on children, of whom 90 per cent were less than 1 year old, duodenal ulcer was recorded only four times. In 566 autopsies on children in a London hospital Paterson [4] encountered two children with peptic ulcer, but he adds that none had been observed in that hospital in the preceding 20 years. Berglund,[1a] reporting one of the largest series, found peptic ulcer 19 times in 1,323 autopsies on children. Among the records of 6,059 autopsies on children under 13 years Guthrie [1c] found nine of peptic ulcer. The increasing frequency with which ulcers are recognized at autopsy is revealed by comparing Holt's figures, cited above, with those of Cole,[1f] who reviewed the autopsies performed in that same hospital during recent years, viz., 1925 to 1950. He found 31 cases among 2,468 autopsies, an incidence of 1.25 per cent.

The coexistence of cerebral and gastroduodenal lesions in infancy and childhood was emphasized over a century ago by Rokitansky.[5] Thereafter this relationship

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Read at the meeting of the American Association of Pathologists and Bacteriologists, Cleveland, April 26, 1951.

From the Departments of Pathology, Ohio State University College of Medicine and Children's Hospital of Columbus.

1. (*a*) Berglund, N.: Acta pædiat. **8**:323, 1929. (*b*) Bird, C. E.; Limper, M. A., and Mayer, J. M.: Ann. Surg. **114**:526, 1941. (*c*) Guthrie, K. J.: Arch. Dis. Childhood **17**:82, 1942. (*d*) Mossberger, J. I.: J. Neuropath. & Exper. Neurol. **6**:391, 1947. (*e*) Ingram, M. D.: Am. J. Roentgenol. **64**:765, 1950. (*f*) Cole, A. R. C.: Pediatrics **6**:897, 1950.

2. Hutchins, L. R.: Northwest Med. **43**:40, 1944.

3. Holt, L. E.: Duodenal Ulcers in Infancy, Am. J. Dis. Child. **6**:381 (Dec.) 1913.

4. Paterson, D.: Lancet **1**:63, 1922.

5. Rokitansky, C.: A Manual of Pathological Anatomy, London, Sydenham Society 1849, vol. 2, pp. 36-37.

was largely ignored, although in 1929 Berglund noted that six of the children with peptic ulcer died of purulent meningitis and that one had hydrocephalus. Cushing [6] stimulated renewed interest in the problem, and there are now in the literature [7] several reports of children with both gastroduodenal and cerebral lesions.

GASTRIC ULCER AND GASTROMALACIA (TABLE 1)

The gastric lesions considered here were all acute. Ingram [1e] recently reported a case of acute gastric ulcer in a child and tabulated 33 others that he found in the literature. Unlike the chronic ulcer, many of the acute lesions are found in the fundus and along the greater curvature. Perforation is not uncommon in children [8] and occurs more frequently in gastric than in duodenal ulcers. The ulcers may be multiple, as in Case 1, where 47 discrete lesions could be counted in the fundus of the stomach.

The distinction between gastric ulcer and gastromalacia may be somewhat arbitrary, but in terms of gross pathologic changes it serves to differentiate the small, sharply circumscribed, punched-out ulcer from a large, often poorly demarcated area of dissolution. The occurrence of numerous microscopic erosions and ulcerations in the immediate vicinity of the area of gastromalacia in Case 2 suggests that the former may, in some instances at least, be a precursor of the latter.

The antemortem or postmortem occurrence of gastromalacia has been argued since the time of Hunter, who maintained that it represented postmortem autolysis. Cruveilhier [9] illustrated the lesion, noted its frequent occurrence in early infancy, and insisted that it is ante mortem. This view was shared by Rokitansky,[5] who also observed that " . . . it occurs both in children and in adults as a sequela of acute affections of the brain and its membranes, and more especially of tubercular meningitis at the base of the brain."

The recent case of Gottlieb, Chu, and Sharlin [7g] offers unequivocal evidence that gastromalacia can occur before death. Their patient, a newborn full-term boy, began passing blood in both stool and vomitus on the second day of life. At operation a blood clot was found plugging a 3 cm. perforation in the stomach. Subsequent autopsy revealed an intracranial hemorrhage.

In both the cases of gastromalacia reported here the lesion had perforated before death, as was manifested by the presence of a hemoperitoneum and peritonitis at autopsy. The danger of perforating the weakened gastric wall by too vigorous

6. Cushing, H. W.: Peptic Ulcer and the Interbrain, in Papers Relating to the Pituitary Body, Hypothalamus, and Parasympathetic Nervous System, Springfield, Ill., Charles C Thomas, Publisher, 1932.

7. (*a*) Masten, M. G., and Bunts, R. C.: Neurogenic Erosions and Perforations of Stomach and Esophagus in Cerebral Lesions: Report of 6 Cases, Arch. Int. Med. **54**:916 (Dec.) 1934. (*b*) Grant, F. C.: Ann. Surg. **101**:156, 1935. (*c*) Opper, L., and Zimmerman, H. M.: Yale J. Biol. & Med. **11**:49, 1938. (*d*) Webster, R.: M. J. Australia **1**:1061, 1938. (e) Hartung, C. A., and Warkany, J.: Duodenal Ulcer as Cause of Death in Case of Meningococcic Meningitis, J. A. M. A. **110**:1101 (April 2) 1938. (*f*) Langlois, M.: Laval méd. **7**:534, 1942. (*g*) Gottlieb, C.; Chu, F., and Sharlin, H. S.: Radiology **54**:595, 1950. Mossberger.[1d]

8. Rosenberg, A. A., and Heath, M. H.: J. Pediat. **28**:93, 1946. Wright, L.; Wright, T., and Scott, B. E.: Ibid. **37**:905, 1950.

9. Cruveilhier, J.: Anatomie pathologique du corps humain, Paris, J. B. Baillière, 1829-1835.

intubation and gavage was pointed out by Heyde and Robinson [10] and may have been a factor in Case 2. In Case 3 there was also ulceration with perforation of the esophagus just above the level of the diaphragm. Similar perforation of an esophageal ulcer during an attack of bulbar poliomyelitis was recently observed in two adults.[11]

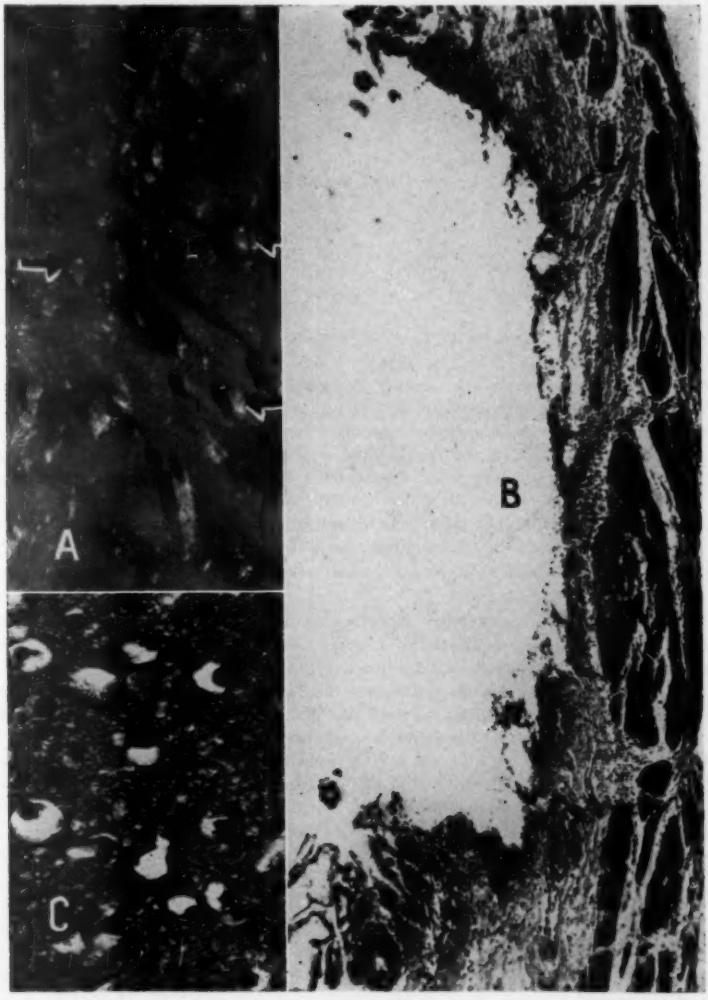

Fig. 1 (Case 1).—*A*, multiple acute ulcers of the gastric mucosa; 16 are present in the area shown. Twice natural size.

B, section of an ulcer showing destruction of the mucosa and most of the submucosa. An inflammatory cell infiltrate of lymphocytes and a few neutrophils is seen in the base and margins. Hematoxylin and eosin stain; ×25.

C, terminal intracellular edema of cortical neurons. Hematoxylin and eosin stain; × 275.

10. Heyde, E. C., and Robinson, S.: Gastroenterology **11**:519, 1948.

11. Erskine, F. A.; Mason, J. H., Jr., and McDade, H.: Am. J. Med. **8**:239, 1950.

CASE REPORTS

CASE 1.—*Clinical Course.*—A white girl, 3 days old at the time of death, was born with the umbilical cord wrapped around her neck. She was difficult to resuscitate and more than an ounce of meconium-like material was aspirated from the trachea. The child never breathed well, and roentgenograms revealed air in the mediastinum and left side of the chest. On tapping, 12 cc. of air was removed from the mediastinum and 50 cc. from the left pleural space. Evidence of progressive cerebral anoxia developed: generalized muscular rigidity; carpopedal spasm; a fixed, staring gaze, and gasping respiration.

Autopsy (CA-51-17, performed five hours after death).—The weight was 3,100 gm. An extensive hemorrhage was present in the thymus and the anterior mediastinum. Although the chest was opened under water, there was no evidence of pneumothorax, nor was air trapped in the areolar tissue of the mediastinum. In each lung the posterior portion of the lower lobe was atelectatic. The stomach contained 7 cc. of partly clotted blood. The mucosa was rugose and bore not less than 47 small ulcers, ranging from 0.5 to 3.0 mm. in diameter and surrounded by a narrow zone of hyperemia (Fig. 1*A*). No blood or ulcers were found in the small or the large intestine. Each adrenal weighed 4 gm. and was grossly normal. The brain weighed 430 gm. (normal, 335 gm.). It was firmer than normal, the convolutions were markedly flattened, and there was a definite pressure groove about the cerebellar tonsils. Sections through the cerebral and cerebellar hemispheres, as well as sections through the brain stem, showed no evidence of hemorrhage.

Microscopic Examination.—In the lungs were the squames and granular debris of aspirated amniotic fluid. The alveoli in some areas were filled with edema fluid and often infiltrated by neutrophils and lymphocytes. The stomach ulcers showed destruction of mucosa, muscularis mucosae, and submucosa; the base was formed by the muscle wall (Fig. 1*B*). In some only the mucosa was absent, the muscularis remaining intact. Inflammatory cell infiltrates, scanty, composed of a few neutrophils and lymphocytes, were present in the edematous tissues in the margin and the base of the ulcers. Throughout the brain there was obvious edema, characterized by accumulation of fluid about the ganglion cells and vessels as well as within the cytoplasm of the glia cells (Fig. 1*C*). The eccentric position of the nucleus in the ganglion cell, pyknosis, and chromatolysis were perhaps due to the associated anoxia.

Final Diagnoses.—Lobular pneumonia, cerebral edema and anoxia, multiple acute gastric ulcers.

CASE 2.—*Clinical Course.*—A 5-day-old premature boy was brought to the hospital with a history of hypothermia and regurgitation of almost every feeding since birth. On admission the infant was slightly jaundiced; a subconjunctival hemorrhage was present in the right eye, and there were many petechiae on the trunk. The abdomen was distended and tympanitic. Blood studies revealed a hemoglobin level of 10.2 gm., a red blood cell count of 2,800,000, a white cell count of 12,200, and a carbon dioxide-combining power (plasma) of 13.2 mEq. per liter. The baby was placed in an incubator with a continuous oxygen supply and given 30 cc. of whole blood by transfusion and sodium bicarbonate intravenously. Buffered crystalline penicillin U. S. P. and parenteral sulfadiazine therapy was begun. The stomach was aspirated frequently, but distention continued to recur. About five hours after admission a small amount of blood-tinged mucus was expelled through the rectum. The patient was given a second transfusion, but his condition continued to deteriorate, and he died 15 hours after admission.

Autopsy (CA-50-86, performed 11 hours after death).—The body length was 38 cm.; the weight, 1,200 gm. When the abdomen was opened, about 20 cc. of blood flowed out; many clots were scattered throughout the peritoneal cavity. The loops of intestine were matted together by a yellow-green plastic exudate. The stomach was collapsed, and in the fundus along the greater curvature the wall was gray and paper thin for an area 2 cm. in diameter. In the center of this was a 13 mm. perforation (Fig. 2*A*). The thin region of gastromalacia was sharply demarcated by a narrow band of hyperemia resembling the zone of reactive hyperemia seen at the margins of an infarct. There was no evidence of ulceration in the mucosa of the small and large intestine, and no blood was present in the lumen. Each of the adrenals weighed 3 gm., the cortices contained no orange-yellow granules (lipids) and the medullae

were semiliquid and brown. When the calvarium was opened, the brain protruded as if under tension. Both cerebral hemispheres were fluctuant, and during removal they ruptured, permitting a large amount of partly clotted blood to escape from the ventricles. Small hemor-

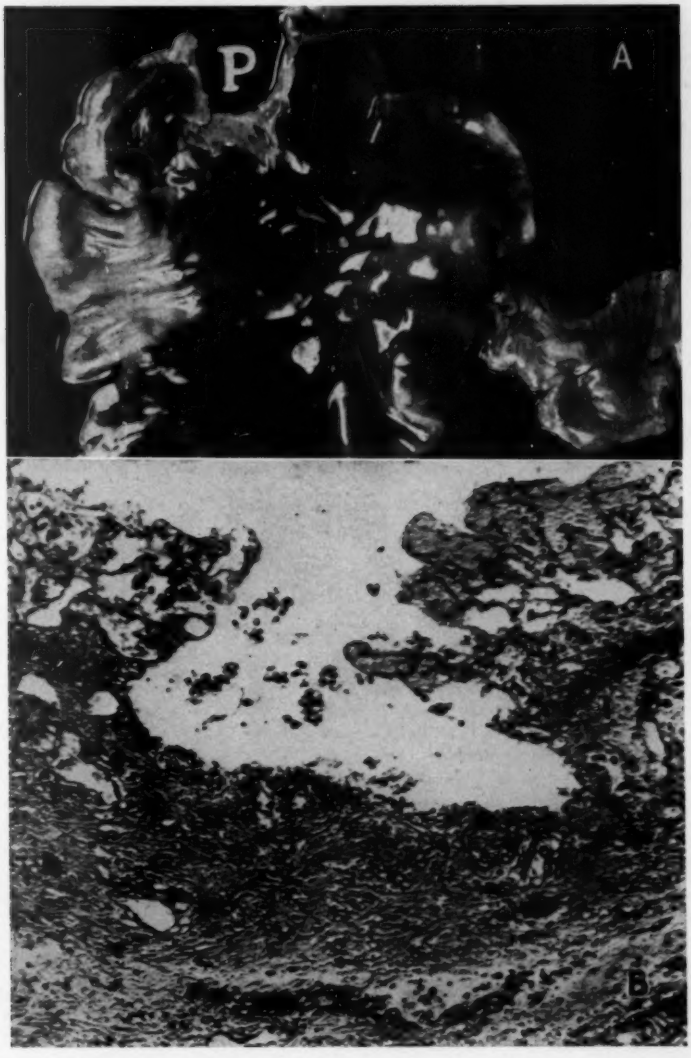

Fig. 2 (Case 2).—*A*, mucosal surfaces of the stomach, esophagus (left), and duodenum (right). Along the upper margin of the stomach is a large U-shaped perforation (*P*) bordered by a narrow pale gray zone of necrotic tissue; beyond this the mucosa is hyperemic. Natural size.

B, one of several acute ulcers not visible grossly found within the area of hyperemia. The capillaries and venules at the mucosal surface immediately adjacent to the ulcer are greatly engorged. The ulcer has penetrated the muscularis mucosae. Hematoxylin and eosin stain; × 120.

rhages were present in the adjacent cerebral tissue as well as in the subarachnoid space about the brain stem and cerebellum.

Microscopic Examination.—The margin of the perforation in the stomach was completely necrotic. Peripheral to this was a broad band of intense hyperemia within which were several small acute ulcers. These penetrated the muscularis mucosae; the base rested in the submucosa and was formed of fibrin and hyalinized connective tissue (Fig. 2*B*). The adjacent tissue was edematous; only an occasional lymphocyte or macrophage was seen. On the peritoneal surface was a fibrinous exudate, the serosa was necrotic, and in the edematous subserosa were macrophages and scattered neutrophils. The adrenals showed preservation of most of the provisional cortex, with only small foci of degeneration. In the brain there were petechiae and degenerative changes in all the paraventricular structures, including the hypothalamus.

Final Diagnoses.—Bilateral intraventricular hemorrhage; gastromalacia with perforation, hemoperitoneum, and peritonitis.

Case 3.—*Clinical Course.*—A 9-year-old white boy was admitted to the hospital with a history of headache, neck pain, and vomiting for three days before admission. He was dehydrated, had difficulty in swallowing, and spoke with a nasal twang. His lips were slightly cyanotic, but respirations were regular, although mainly abdominal. The patient was given intravenous fluids and placed in an oxygen tent. Persistent suction for removal of tracheobronchial mucus was necessary. Twenty hours after admission he vomited "coffee-ground" material; he was given a blood transfusion and placed in a respirator. However, 45 minutes later he had a

Table 1.—*Cases of Coexistence of Gastric and Cerebral Lesions*

Case	Brain Lesion	Gastromalacia	Gastric Ulcer	Race	Sex	Age	Comment
1	Edema and anoxia	..	+	W	F	3 days	Multiple ulcers in fundus
2	Ventricular hemorrhage	+	+	W	M	6 days	Perforation, hemoperitoneum
3	Poliomyelitis	+	..	W	M	9 yr.	Perforation, esophageal ulcer, hemoperitoneum

convulsion and died. The clinical impression was bulbar poliomyelitis with hemorrhage from Cushing's ulcer.

Autopsy (CA-50-90, performed seven hours after death).—In the left upper quadrant of the abdomen was a retroperitoneal hemorrhage; blood clots were also present on the peritoneal surfaces of the stomach and the spleen. Numerous fibrinous adhesions extended between the stomach and the parietal peritoneum. The fundus of the stomach bore a 2.5 cm. perforation along its greater curvature (Fig. 3*A*). In the thin portion of the wall adjacent to the perforation were several small hemorrhages. No additional ulcers were visible on the mucosal surface. The visceral and parietal pleura were smooth and glistening except in the region of the lower lobe of the left lung, where they were greenish brown and covered with a layer of fibrin. Just above the level of the diaphragm the wall of the esophagus was perforated by a ragged, irregular opening 4 mm. in diameter. The adjacent esophageal mucosa was ulcerated and hemorrhagic (Fig. 3*A*). Although some partly digested blood was present in the stomach there was none in the small or the large intestine. The adrenals were large, each weighing 7 gm., and the cortices were rich in lipids. The medullae were firm and brown. The brain weighed 1,510 gm. The convolutions were flattened; the arachnoid and brain substance were hyperemic.

Microscopic Examination.—The lower lobe of the left lung in the region that showed gross evidence of the action of gastric juice was the seat of extensive interstitial and intra-alveolar hemorrhage. The area of malacia and perforation in the stomach and esophagus was necrotic, the peripheral zone edematous, and the capillovenous bed engorged. Inflammatory cells were few. The brain stem showed acute swelling of the ganglion cells in many regions (Fig. 3*B*). Perivascular collections of lymphocytes were prominent in these areas.

Final Diagnoses.—Bulbar poliomyelitis, gastromalacia, perforation of the stomach and esophagus, and hemoperitoneum.

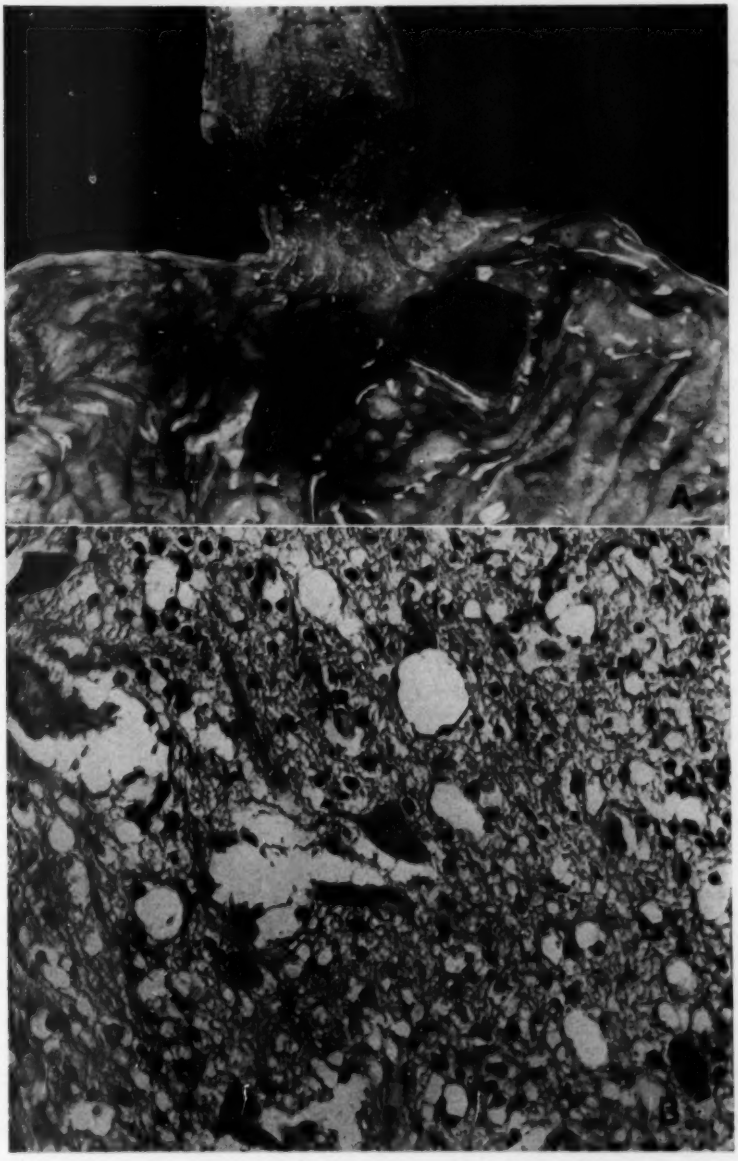

Fig. 3 (Case 3).—*A*, mucosal surface of the esophagus and fundus showing a large perforation along the greater curvature of the latter and hemorrhagic ulceration of the esophagus. Natural size.

B, interstitial and intracellular edema in brain stem. Note karyolysis and chromatolysis with eccentric nuclei and central loss of tigroid substance in ganglion cells. Hematoxylin and eosin stain; × 300.

DUODENAL ULCER (TABLE 2)

All reported series of cases of peptic ulcer in infants and children show a marked preponderance of duodenal over gastric ulcers. The ratios vary from 2:1 to 6:1[12]; in the present group of cases it approximated 2:1. With increasing clinical awareness that these lesions occur in the very young, they are being diagnosed oftener, and early enough that surgical intervention may be successful.[13]

Perforation and hemorrhage are frequent complications.[14] In this series, none of the duodenal ulcers perforated into the peritoneal cavity, but three had eroded the pancreas, and three others had penetrated to the serosa. In three cases the ulcer was the source of massive intestinal hemorrhage (Table 2). All ulcers were proximal to the ampulla of Vater, usually within a few millimeters of the pylorus. In six instances only a single ulcer was found; in two the ulcers were multiple.

Histologically, the ulcers were acute, with a scanty inflammatory cell infiltrate and some edema and hyperemia of the adjacent tissues. In several instances thrombi were present in the small vessels of the submucosa, suggesting that the ulcers arose as small infarcts in the mucosa which sloughed out, exposing the underlying tissues to the action of the digestive juices. This interpretation loses some of its force because of the difficulty of deciding whether the thrombosis occurred before or after inception of the ulcer.

In each case reported here cerebral symptoms were a prominent part of the clinical picture, and a brain lesion was found at autopsy.

CASE REPORTS

CASE 4.—*Clinical Course.*—A 6-month-old girl was admitted with a history of diarrhea for four days. Three months before this she had been hospitalized for a week with virus pneumonia but was still coughing on discharge. The father was absent without leave from a tuberculosis sanatorium. On admission she weighed 9 lb. (4,082 gm.). She had a temperature of 103 F. and enlarged lymph nodes in the neck and the abdomen. The spinal fluid showed 238 white blood cells per cubic millimeter, of which 34 per cent were neutrophils and 66 per cent were lymphocytes; the total protein was 55 mg. per 100 cc. The tuberculin test was positive. Under treatment the diarrhea subsided three days after admission; however, a day later the anterior fontanelle was found to be bulging, the head retracted, and the back arched. The infant failed rapidly and died on the 11th hospital day.

Autopsy (CA-49-116, performed four hours after death).—The lungs were the seat of tuberculous pneumonia in the right middle and both lower lobes; miliary tubercles were scattered throughout the remaining lung parenchyma. The hilar, para-aortic, and mesenteric lymph nodes were enlarged and caseous. The serosa of the intestine was dotted by tubercles, and tuberculous ulcers were present in the terminal ileum and caecum. In the mucosa of the duodenum, 5 mm. beyond the pylorus, was an irregular shallow 1 cm. ulcer accompanied by two satellite erosions, each 3 mm. in diameter. There was no evidence of tuberculosis in the adjacent tissues. Each adrenal weighed 2 gm.; the cortex was narrow and yellow, the medulla firm and gray. The brain weighed 750 gm., was soft in consistency, and without evidence of ventricular dilatation. At the base, particularly about the brain stem and the optic chiasm, was a thick grayish-yellow exudate. Many small tubercles were present in the leptomeninges over the temporal and parietal lobes.

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12. Donovan, E. J., and Santulli, T. U.: Gastric and Duodenal Ulcers in Infancy and Childhood, Am. J. Dis. Child. **69**:176 (March) 1945.

13. (*a*) Plummer, G. W., and Stabinus, S. J.: J. Pediat. **37**:899, 1950. (*b*) Bird and others.[1b]

14. Margolis, B.; Valdes-Dapena, M., and Boles, R. S.: Gastroenterology **12**:489, 1949. Plummer and Stabinus.[13a]

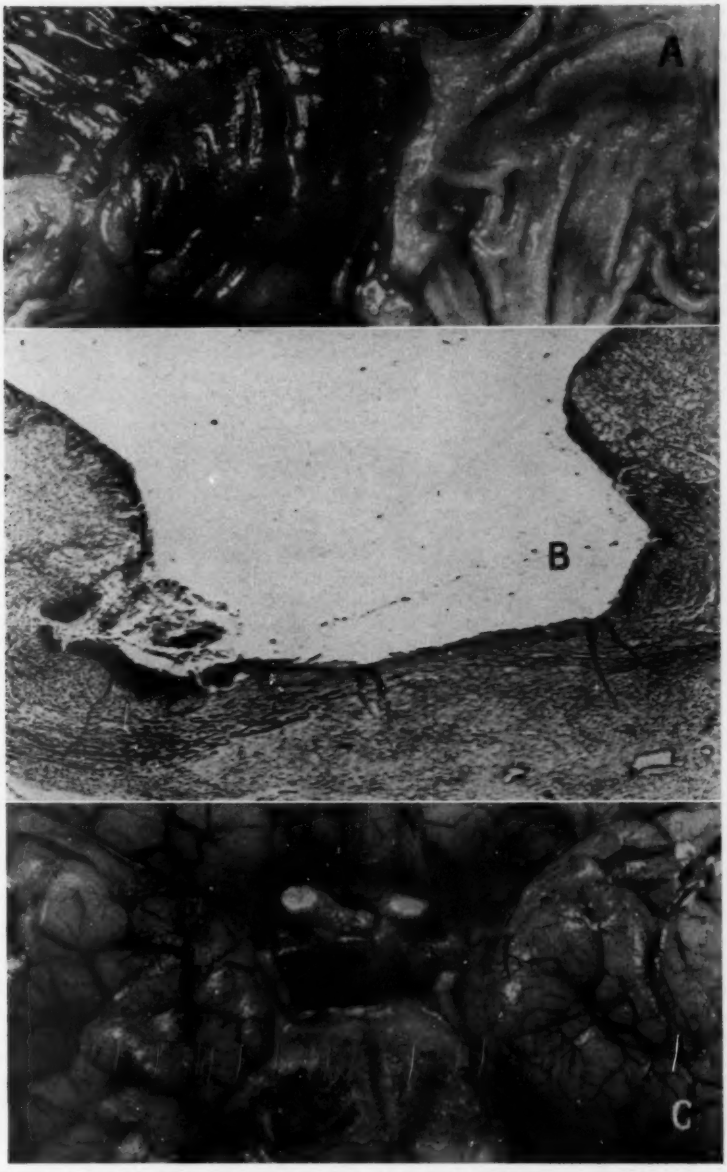

Fig. 4 (Case 5).—*A*, punched-out ulcer in the duodenal mucosa, 1 cm. beyond the pylorus. Note the surrounding zone of hyperemia. One and a half times natural size.

B, section of ulcer showing partial penetration of circular layer of smooth muscle and almost complete absence of inflammatory response. Hematoxylin and eosin stain; × 25.

C, base of the brain in the region of the optic chiasm and pons covered by a thick plastic exudate. One and a half times natural size.

Microscopic Examination.—Miliary tubercles were found in tissues of the following organs: lungs, liver, spleen, kidneys, ileum, cecum, peritoneum, and lymph nodes. The wall of the duodenum was penetrated by a large ulcer, which was associated with little inflammation and was without evidence of tuberculosis. The base of the ulcer was formed by the pancreas. The satellite ulcers appeared as small erosions of the mucosa; they did not penetrate and were unaccompanied by inflammatory changes. There were tubercles within the brain stem and the stalk of the pituitary, as well as in the leptomeninges.

Final Diagnosis.—Tuberculous pneumonia with miliary dissemination, tuberculous meningoencephalitis, and acute nontuberculous duodenal ulcer.

Case 5.—*Clinical Course.*—A 33-month-old Negro boy was well until three months before death; then he became feverish, complained of abdominal pain, and had a convulsion. On admission he was still fairly well nourished but appeared acutely ill. A roentgenogram of the chest showed miliary infiltration; the tuberculin test was positive. Streptomycin therapy was begun; 150 mg. was injected intramuscularly every four hours for 56 days and 25 mg. intrathecally daily for 12 days, then on alternate days until 900 mg. had been given. Streptomycin therapy was discontinued because of the absence of any noticeable clinical response. Thiazolsulfone (promizole® [4,2′-diaminophenyl-5′-thiazolylsulfone]) was administered in doses of 0.25 gm. twice daily for 16 days before death. During the last 10 days of life the child was comatose; death occurred on the 82d hospital day.

Autopsy (CA-49-103, performed three hours after death).—The lungs were studded with miliary tubercles, but neither a Ghon tubercle nor markedly enlarged and caseating hilar lymph nodes were found. On the posterior wall of the duodenum, 6 mm. below the pylorus, was a deep mucosal ulcer, 5 mm. in diameter, surrounded by a 2 mm. broad zone of hyperemia (Fig. 4*A*). There was no evidence of hemorrhage, and the rest of the gastrointestinal tract was normal. Each adrenal weighed 2 gm.; the cortex was orange-yellow, the medulla firm and brown. The brain weighed 1,100 gm. Surrounding the infundibulum and optic chiasm was a firm yellow-gray exudate (Fig. 4*C*) that extended posteriorly to occlude the foramina of Magendie and Luschka. There was considerable dilatation of the lateral and third ventricles, producing an internal hydrocephalus.

Microscopic Examination.—Miliary tubercles were scattered throughout the lungs, but were also found in the liver, spleen, kidneys, and hilar lymph nodes. A striking feature of the tubercles was the extensive fibrosis they had undergone and the small amount of caseation necrosis that was present. This probably may be attributed to the streptomycin therapy which the patient had received. The ulcer of the duodenum had destroyed the mucosa and Brunner's glands. The base was formed by the necrotic and partly hyalinized circular layer of smooth muscle (Fig. 4*B*). The longitudinal muscle bundles and the serosa were edematous and infiltrated by a few macrophages and eosinophils, with only a rare neutrophil and lymphocyte present. In sections of the brain the tuberculous meningitis was accompanied with a fibrosis of the tubercles that was similar to, if less impressive than, that noted in the lungs. Small inactive-appearing tubercles were also present in the wall of the third ventricle.

Final Diagnoses.—Miliary tuberculosis with tuberculous meningoencephalitis and an acute nontuberculous duodenal ulcer.

Case 6.—*Clinical Course.*—A white boy of 4 years first became ill two months prior to admission, with headache, painful knees, and vomiting. He recovered and appeared well until nine days before admission; then these symptoms returned. Physical examination showed a child chronically ill, with an enlarged liver, moderate nuchal rigidity, and fever. Roentgenograms of the chest revealed bilateral hilar shadows and probable miliary infiltration of the lung fields. The boy's reflexes were hyperactive, and the Babinski, Gordon, and Kernig signs were all present. The spinal fluid showed 230 white blood cells per cubic millimeter; 48 per cent were neutrophils and 52 per cent were lymphocytes; the total serum protein was 137 mg. per 100 cc. Despite supportive therapy the child died on the 10th hospital day.

Autopsy (CA-50-15, performed four hours after death).—The lungs showed an irregular 2 cm. area of caseation necrosis in the left upper lobe and tiny miliary tubercles scattered throughout the other lobes. The hilar lymph nodes were enlarged and the seat of caseation necrosis. The spleen and the liver bore many subcapsular and parenchymal tubercles. On the posterior mucosal surface of the duodenum, 4 mm. below the pylorus, was a sharply demarcated, "punched-

out" 5 mm. ulcer (Fig. 5*A*). The remainder of the gastrointestinal tract was normal. Each adrenal weighed 3 gm. and appeared grossly normal. The cortices contained bright orange-yellow granules; the medullae were firm and brown. The brain weighed 1,225 gm. At its base, particularly about the optic chiasm and extending over the pons and onto the medulla, was a yellow-gray exudate. Sections through the cerebral hemispheres revealed a moderate hydrocephalus.

Microscopic Examination.—The lungs were the seat of tuberculous pneumonia with miliary dissemination to the surrounding parenchyma as well as to the spleen, the liver, and lymph nodes. The ulcer in the duodenum had destroyed mucosa and submucosa, but the layers of smooth muscle were intact (Fig. 5*B*). The base was covered with a layer of fibrin containing

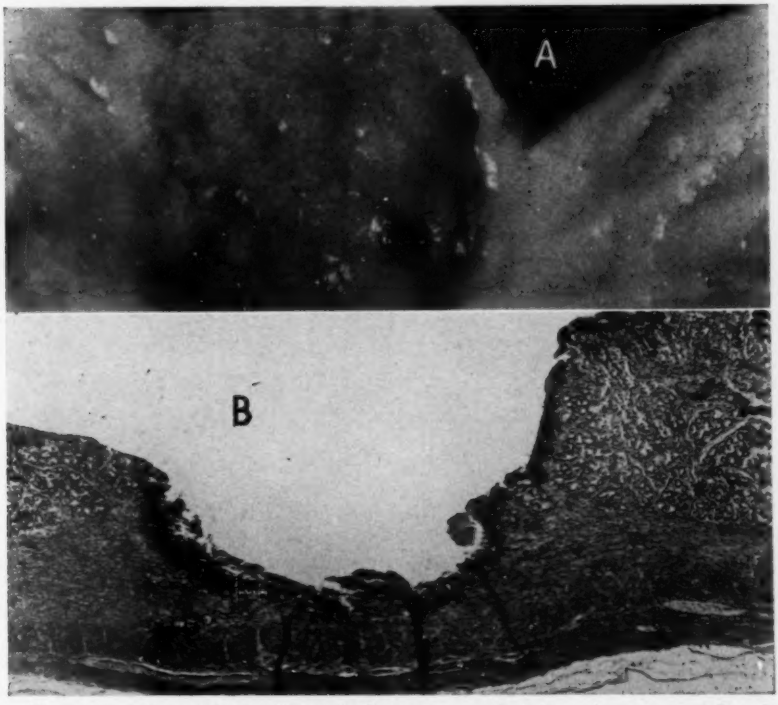

Fig. 5 (Case 6).—*A*, in the dilated duodenum is a small shallow ulcer, 4 mm. below the pylorus. Three times natural size.

B, section of ulcer showing destruction of mucosa and submucosa, but intact layers of smooth muscle. The base is covered by a layer of fibrin. Hematoxylin and eosin stain; × 20.

scattered neutrophils and occasional lymphocytes. Histologic sections of the brain confirmed the gross finding of tuberculous meningitis and in addition disclosed an encephalitis involving particularly the brain stem and manifested by perivascular cuffs of lymphocytes. A large tubercle was present within the brachium conjunctivum.

Final Diagnoses.—Primary pulmonary tuberculosis with miliary dissemination, tuberculous meningoencephalitis, and acute nontuberculous duodenal ulcer.

CASE 7.—*Clinical Course.*—A boy, 14, was well until two weeks before death, when he complained of generalized malaise and anorexia. The symptoms of bulbar poliomyelitis gradually developed, prominent among which were difficulty in speech and swallowing. On admission he

was given 300,000 units of buffered crystalline penicillin U. S. P. daily, subsequently increased to 600,000 units. Later this was supplemented by 125 mg. of streptomycin every three hours to treat an otitis media. On the fourth hospital day the patient's breathing became irregular and he was placed in a respirator. On the sixth day, 24 hours before death, he twice vomited brown fluid. His temperature "spiked" to 109 F., but dropped to 100 F. after ice enemas and tepid sponges. Shortly before death his lips and conjunctivas became very pale and he passed several tarry stools.

Autopsy (CA-49-96, performed 10 hours after death).—The stomach was distended by 400 cc. of "coffee-ground" material; the mucosa was rugose, free of ulceration. In the duodenum, 1 cm. below the pylorus, was an irregular deep ulcer that measured 2.5 by 1.5 by 0.5 cm. The floor of the ulcer was formed by the pancreas; near one margin was an eroded blood vessel, from which bleeding probably occurred. Grouped about this ulcer for a distance of 5 cm. were

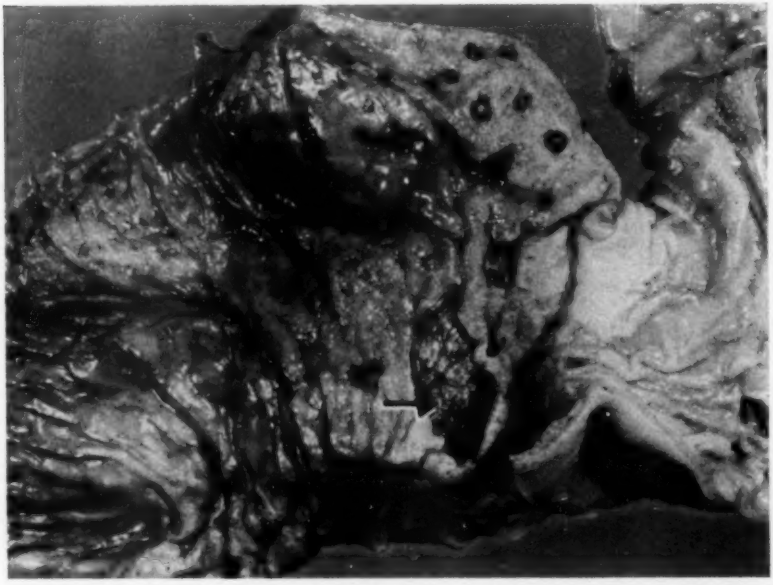

Fig. 6 (Case 7).—Pylorus and duodenum; the latter exhibits many deep, sharply circumscribed ulcers. The largest lies 1 cm. beyond the pylorus (arrow); it has laid bare the underlying pancreas. Nearby are several smaller ulcers, irregular or circular in outline. Natural size.

11 additional ulcers ranging in size from 3 to 7 mm. (Fig. 6). Both the small and the large intestine were almost filled with partly digested blood, but no additional ulcers were found. Both adrenals were of normal size and shape. The cortices were bright orange-yellow; the medullae, firm and gray. The brain weighed 1,750 gm., the arachnoid was thickened and hyperemic, and the cerebral convolutions were flattened. On section the brain was diffusely hyperemic.

Microscopic Examination.—The ulcers in the duodenum were all acute; several penetrated the wall, and the underlying pancreas was partly digested. Scanty infiltrates of neutrophils and lymphocytes were present in the edematous margins. The brain and cord showed evidence of poliomyelitis. The anterior horn cells of the cervical cord were absent or degenerated; similar changes were found in the ganglia of the brain stem. Perivascular collections of lymphocytes were not a prominent feature, although present in several fields.

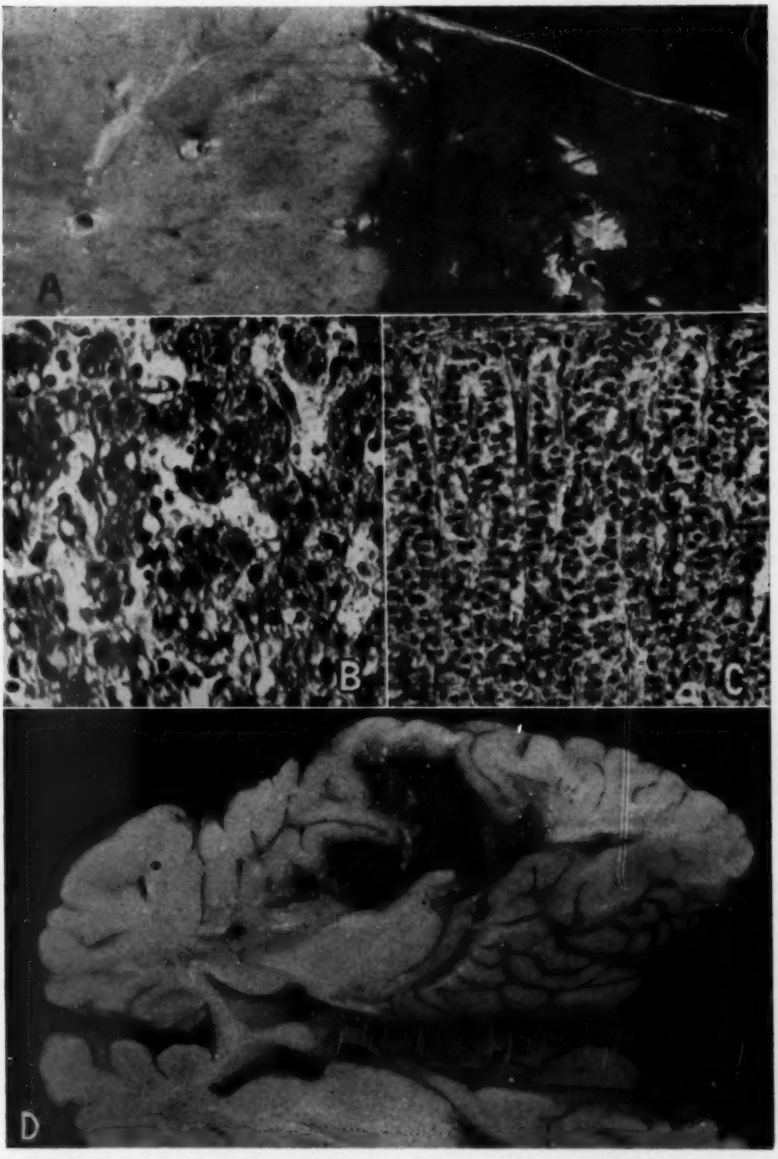

Fig. 7 (Case 8).—*A*, cut surface of liver showing rather sharp demarcation between the pale yellow-brown area and the darker, grossly normal liver tissue. Twice natural size.

B, section of liver showing fat in the liver cells of the pale area (lower left) and normal cells from the dark area (upper right). Hemotoxylin and eosin stain; × 275.

C, normal adrenal cortex. Hematoxylin and eosin stain; × 105.

D, brain showing massive hemorrhage in the right cerebral hemisphere with rupture into lateral ventricle.

Final Diagnoses.—Bulbar poliomyelitis, multiple acute duodenal ulcers, gastrointestinal hemorrhage.

CASE 8.—*Clinical Course.*—A 2-month-old white boy was admitted with a history of food allergy. He had suffered a convulsion 10 days before, which was successfully treated with calcium. Thereafter his hands were always clenched and his arms rigidly held in flexion. On the day before admission a discharge from the right ear was first noted. Physical examination revealed a poorly nourished, acutely ill infant with discrete pleomorphic crusting lesions and a background of erythema that covered the entire body. Pus drained from each ear; on culture it yielded Proteus vulgaris and a nonhemolytic Staphylococcus aureus. The tongue and both eyes deviated to the right. By the third hospital day the dermatitis had improved in consequence of topical applications, and the neurological findings were less evident. Shortly before death the temperature rose to 103 F., and the infant died unexpectedly on the sixth hospital day.

Autopsy (CA-49-127, performed six hours after death).—The liver weighed 200 gm.; the surface was smooth, the capsule thin and transparent. On the dorsum was a rather sharply demarcated large irregular area that was pale orange-yellow as compared with the red-brown color of the adjacent liver tissue. On section this discoloration extended through the parenchyma (Fig. 7*A*). The duodenum bore a 5 mm. shallow ulcer 3 cm. below the pylorus and just proximal to the papilla of Vater. The ulcerated mucosa was sharply elevated by the underlying pancreas. Each adrenal weighed 2 gm. The cortices contained orange-yellow granules; the medullae were firm and brown. Inspection of the cranium revealed pus in the petrous portion of the right temporal bone, but no epidural or subdural abscess was present. There was thrombosis of the right lateral and cavernous sinuses as well as of the superior longitudinal sinus; thrombi from the latter extended into the right bridging veins. In the right cerebral hemisphere a massive hemorrhage had destroyed much of the centrum and had ruptured into the right lateral ventricle (Fig. 7*D*). No suppurative lesions could be demonstrated.

Microscopic Examination.—The duodenal ulcer had penetrated into the submucosa, where the base was formed by radicals of the accessory duct of Santorini and aberrant pancreatic acinar tissue. Below the latter lay the normal layers of smooth muscle. About the margins and in the base were scant infiltrates of plasma cells, lymphocytes, and occasional neutrophils. The liver was the seat of marked fatty dystrophy in the region that appeared grossly yellow; much less fat was present in the liver cells from other areas (Fig. 7*B*). The adrenal cortex was well preserved; the cells were finely vacuolated and free of degenerative changes (Fig. 7*C*). The medulla was normal. The brain was edematous, with hemorrhage and necrosis of the centrum and the right paraventricular tissue. Thrombi were present in several of the subarachnoid veins as well as in the dural sinuses.

Final Diagnoses.—Acute duodenal ulcer, otitis media, right dural and longitudinal sinus thromboses, cerebral hemorrhage.

CASE 9.—*Clinical Course.*—The patient was a mentally retarded white boy, 12 years old, who had been cyanotic since birth. On admission he appeared fairly well nourished but with a generalized dusky color, cyanosis of the lips, and clubbing of the fingers and toes. A loud systolic murmur was heard over the entire precordium. The hemoglobin level was 22.8 gm.; the red blood cell count, 6,700,000; the oxygen saturation of the arterial blood, 50.5 per cent; the oxygen saturation of the venous blood, 22.8 per cent, and the carbon dioxide-combining power (plasma), 10.6 mEq. per liter. The electrocardiogram with standard limb leads showed high voltage in P-2, sinus tachycardia, and strong right axis deviation. Four days before death angiocardiography was carried out under anesthesia induced with sterile thiopental sodium U. S. P. Immediately after the injection of 35 cc. of iodopyracet concentrated solution U. S.P., the patient became deeply cyanotic, the pulse rate increased to 140, and the heart sounds became faint. This was followed by a five minute interval of cardiac arrest, during which the heart was manually activated; nevertheless, the patient remained almost blue-black in color. After administration of phenylephrine hydrochloride injection U.S.P. and 100 per cent oxygen the heart began to beat spontaneously, and the patient's condition gradually improved to the point that he could be removed from the operating room. He never regained consciousness and died four days later. During this period the body temperature varied from 103 to 104 F. and the pulse rate from 110 to 130. The clinical impression was tetralogy of Fallot with severe cerebral anoxia and possible vascular thrombosis.

Autopsy (CA-50-103, performed four hours after death).—The heart weighed 145 gm. (normal, 124 gm.); the right ventricular wall was hypertrophied. The pulmonary valve was deformed; both the valve and the conus were stenosed. The ascending aorta was wide and shifted to the right, straddling a 2 cm. interventricular septal defect. In the duodenum, 1 cm.

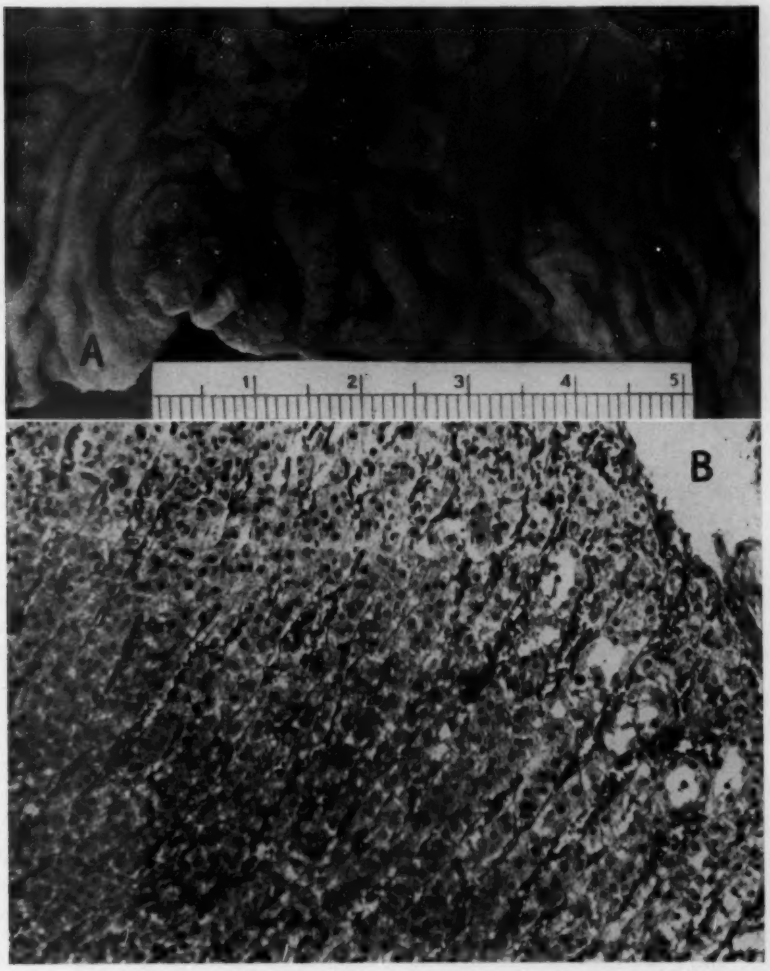

Fig. 8 (Case 9).—*A*, pylorus and duodenum. Near the center of the photograph, 1 cm. beyond the pylorus, is a large ulcer. The base was greenish yellow and moderately firm.
B, moderate hyperplasia of the adrenal cortex, particularly of the fascicular layer. The cells contain only small quantities of lipids. Hematoxylin and eosin stain; × 135.

distal to the pylorus, was a deep ulcer, 8 mm. in diameter. The margins were sharp; the base, greenish yellow and moderately firm (Fig. 8*A*). Two eroded small vessels, representing probable bleeding points, could be identified. The stomach appeared normal; the small and the large intestine were filled with liquid blood. Each adrenal weighed 7 gm.; the cortices

contained orange-yellow granules; the medullae were firm and gray. Permission for examination of the brain was refused.

Microscopic Examination.—The duodenal ulcer had penetrated the submucosa and part of the smooth muscle layer. In the base were several large exposed nerve bundles and a thin film of fibrin. The adjacent tissue was edematous and infiltrated by a small number of lymphocytes, eosinophils, and occasional neutrophils. The adrenal cortex, particularly the fascicular layer, appeared wider than normal; the cells were large, eosinophilic, and contained few lipids (Fig. 8 *B*). Both cortical and medullary cells showed no degenerative changes.

Final Diagnoses.—Acute duodenal ulcer with intestinal hemorrhage, tetralogy of Fallot, and severe cerebral anoxia (clinical).

CASE 10.—*Clinical Course.*—A 32-month-old white girl showed definite mental retardation, manifested by inability to sit up, feed herself, or talk. Born after eight months' gestation, she presented early evidence of cranial synostosis, for which operations were carried out when she was 18 and 21 months old. She had regurgitated her feedings for three days before entering the hospital, but there was no diarrhea; the family physician made a diagnosis of pneumonia. On admission the child appeared microcephalic, malnourished, and dehydrated. Despite supportive therapy she died three days after entering the hospital.

Autopsy (CA-51-5, performed six hours after death).—The body weighed only 8.8 lb. (3,991 gm.) and was 62 cm. in length (normal, 86 cm). There was an oblique deformity of the trunk, also thoracic kyphosis, due to the patient's habit of invariably lying on her right side. In the duodenum, 1 mm. below the pylorus, was an irregular shallow ulcer, measuring 5 by 7 mm. (Fig. 9 *B*). The gastric mucosa was covered by a layer of bloody mucus but was free of ulceration; the small and the large intestine were normal. Each adrenal weighed 1 gm.; the cortices were orange-yellow, the medullae firm and brown. The head was small (circumference, 34 cm.), as were also the eyes and the jaw. The calvarium showed evidence of previous operations for relief of the synostosis, but these were followed by spontaneous fusion. The brain weighed only 240 gm. (normal, 1,060 gm.) but was well proportioned (Fig. 9*A*). On section the cortex and the centrum of the cerebral hemispheres were markedly narrowed; the basal ganglia appeared to be of normal size.

Microscopic Examination.—The lungs were the seat of interstitial pneumonia with edema and atelectasis. The lesion in the duodenum represented an early stage of an acute ulceration. Although all of the mucosa in the affected area was necrotic, portions of it had not sloughed out. Thrombi were found in several of the vessels in the submucosa (Fig. 9 *C*). Inflammatory cells were represented by only an occasional plasma cell and lymphocyte. The brain showed hypoplasia of the cells in the granular layer of the cerebellum and degeneration of the Purkinje cells. There was disorganization of the cerebral cortex with siderosis of some of the ganglion cells and arteriolar walls. Acute swelling of the neurons was found in the brain stem ganglia. The retinas of both eyes were hypoplastic.

Final Diagnoses.—Interstitial pneumonia, acute duodenal ulcer, microcephaly, craniosynostosis, microphthalmos, micrognathia.

CEREBRAL LESIONS (TABLE 3)

In each of the 10 cases reported the cerebral lesion played an important part in the patient's illness; whereas, the peptic ulcer was often an incidental postmortem finding (Tables 1 and 2).

The role of the patient's sex is difficult to evaluate. Although ulcers were more than two times as frequent in males as in females, this must be weighed against the fact that cerebral lesions were almost twice as frequent in males (Table 3). Cole [1f] found a 1:1 ratio of male to female patients; Bird and associates,[1b] a ratio of 1.5:1, compared with 2.3:1 in the present series.

Tuberculous meningoencephalitis was associated with a duodenal ulcer in three instances; the patients were 6 months, 33 months, and 4 years old. In all there was primary pulmonary tuberculosis with miliary spread. A thick gray exudate was always present about the base of the brain, particularly in the region of the optic

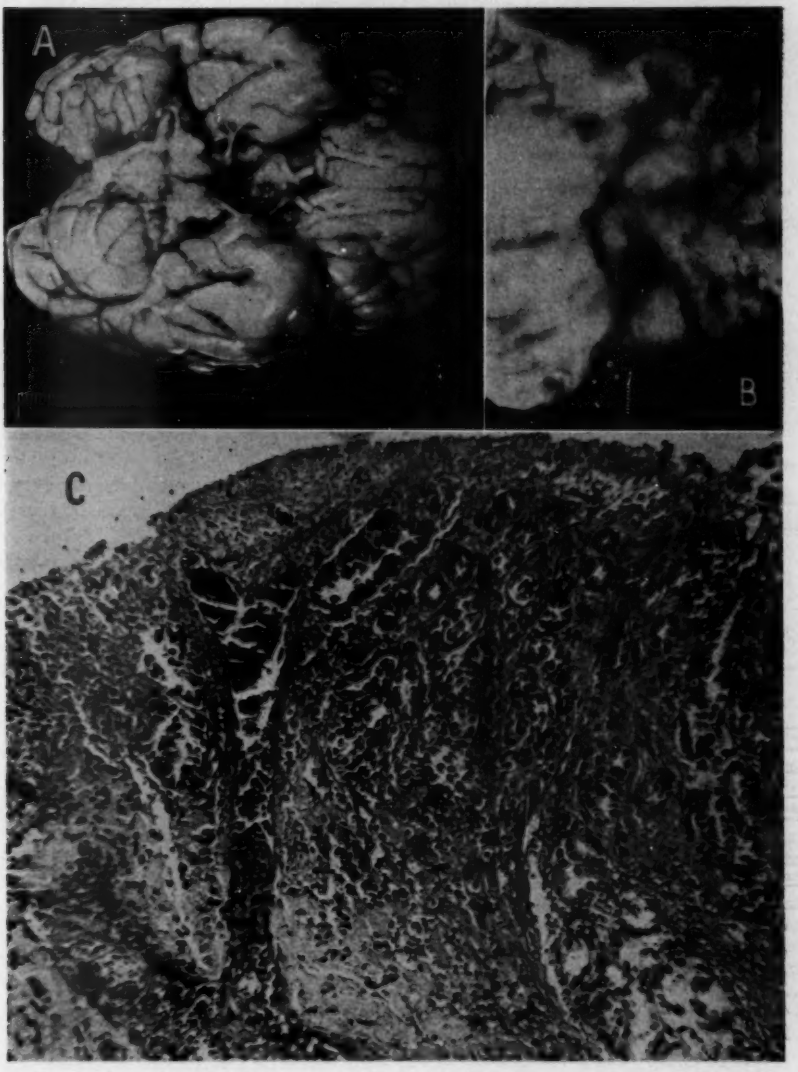

Fig. 9 (Case 10).—*A*, inferior aspect of brain showing its very small size (240 gm.) but almost normal proportions.

B, pylorus and duodenum. An irregular shallow ulcer is seen 1 mm. below the pylorus. Twice natural size.

C, base of the ulcer with preservation of some glandular epithelium. Thrombosis of vessels is seen in the submucosa and in the mucosa. There are fibrinoid degeneration of the connective tissue and scanty inflammatory cell infiltration. Hematoxylin and eosin stain; × 120.

chiasm and infundibulum. Encephalitis was manifested by perivascular cuffs of lymphocytes, particularly in the brain stem. Discrete tubercles were found within the brain stem and the pituitary stalk (Case 4), the brain stem and the wall of the third ventricle (Case 5), and the brachium conjunctivum (Case 6). Only in Case 5 did the patient receive streptomycin therapy, and histologically the lesions in the brain and other viscera were characterized by a degree of fibrosis and an absence of caseation necrosis not seen in the others. These three patients with duodenal ulcer were found in a total of 10 with tuberculous meningoencephalitis (Table 3), an incidence of 30 per cent. This confirms Rokitansky's early observation of the frequent coexistence of these two lesions.[8]

TABLE 2.—*Cases of Coexistence of Duodenal and Cerebral Lesions*

Case	Brain Lesion	Race	Sex	Age	Comment
4	Tuberculosis	W	F	6 mo.	3 duodenal ulcers
5	Tuberculosis	C	M	33 mo.	Streptomycin therapy
6	Tuberculosis	W	M	4 yr.	Moderate postinfection hydrocephalus
7	Poliomyelitis	W	M	14 yr.	12 duodenal ulcers, gastrointestinal hemorrhage
8	Sinus thrombosis	W	M	2 mo.	Fatty liver
9	Anoxia	W	M	12 yr.	Tetralogy of Fallot, gastrointestinal hemorrhage, brain not examined
10	Microcephaly	W	F	32 mo.	Bloody mucus in stomach

TABLE 3.—*Incidence of Coexistence of Cerebral Lesions and Gastroduodenal Ulcers in 251 Consecutive Autopsies*

Brain Lesion	Patients			No. with Coexistent Gastroduodenal Ulcers		
	Male	Female	Total	Male	Female	Total
Hemorrhage	16	9	25	1	..	1
Tuberculosis	7	3	10	2	1	3
Edema and anoxia	5	5	10	1	1	2
Meningitis (nontuberculous)	5	4	9	..	..	..
Poliomyelitis	5	3	8	2	..	2
Congenital anomaly	5	3	8	..	1	1
Hydrocephalus	4	2	6	..	..	..
Kernicterus with erythroblastosis	4	1	5	..	..	..
Kernicterus without erythroblastosis	..	3	3	..	..	..
Brain tumor	2	1	3	..	..	..
Sinus thrombosis	1	1	2	1	..	1
Total	54	35	89	7	3	10

Poliomyelitis, bulbar in type, was once associated with gastromalacia and once with multiple duodenal ulcers. In both cases the gastrointestinal lesion was probably the immediate cause of death. In the first instance (Case 3) this was due to perforation of the stomach, with hemoperitoneum and peritonitis; in the second (Case 7), to massive gastrointestinal hemorrhage from the largest of many duodenal ulcers. Evidence of brain stem ganglion cell degeneration was found in both, but was more severe in Case 7, where the process was so acute that little perivascular accumulation of lymphocytes had taken place. These two cases occurred among eight children who died of bulbar poliomyelitis (Table 3); it is interesting to note that of eight adults who died with this disease and came to autopsy during the same period, none showed evidence of gastrointestinal ulceration. However, Heyde and Robinson [10] reported multiple gastroduodenal ulcers in a man, 26, and Erskine and co-workers [11] found esophageal perforations in a woman, 20, and a man, 27; all died with bulbar poliomyelitis.

Meningitis due to organisms other than Mycobacterium tuberculosis, including Eberthella coli, Neisseria intracellularis (N. meningitidis), Diplococcus pneumoniae, and Hemophilus influenzae, was not associated with a gastroduodenal ulcer, although several authors have reported such a coincidence.[15]

The commonest intracranial lesion among the 251 infants and children was brain hemorrhage. In most of the cases this was due to birth trauma, but also included were those in which it was associated with leukemia, aneurysm and accidental injury. The cases so listed number 25 (Table 3), yet in only one (Case 2) was there a gastrointestinal lesion—in this instance, gastromalacia with perforation, hemoperitoneum, and peritonitis. The cerebral injury, due to birth trauma, had produced massive bilateral intraventricular hemorrhage. Petechiae and foci of degeneration were present in all paraventricular structures including the hypothalamus. A similar case was reported by Masten and Bunts [7a] and more recently by Gottlieb and associates.[7g]

Sinus thrombosis with secondary cerebral hemorrhage occurred twice and in one instance (Case 8) was associated with a small duodenal ulcer. In this case the thrombosis of the right cavernous and lateral sinuses and of the superior longitudinal sinus was less striking than was the massive intracerebral hemorrhage it had precipitated. The large, sharply demarcated area of fatty dystrophy in the liver is of interest because of a widespread impression among pathologists that areas of pallor in the liver may indicate the presence of a cerebral lesion. Wenger [16] pointed out that some are the result of local ischemia, while others, as in this instance, are due to the accumulation of fat in the liver cells. How a cerebral lesion might affect the liver in this manner is not clear. That such a relationship may exist is shown in Wilson's disease, characterized by destructive changes in the brain, especially in the putamen, and cirrhosis of the liver.

Cerebral edema and anoxia are often coexistent and are frequently observed in infants, particularly the newborn. Only those cases in which they played a prominent clinical role and were conspicuous features at autopsy are included in the 10 cases listed (Table 3). One patient (Case 1) in this group bore 47 acute gastric ulcers, and another (Case 9) bled from a large duodenal ulcer. In the former, a 3-day-old infant, the cerebral edema and anoxia were associated with partial asphyxia due to impairment of pulmonary aeration, which had been present since birth. Histologically there were diffuse cerebral edema and the so-called acute swelling of the ganglion cells found in anoxia. The other patient mentioned died without regaining consciousness after a 15 to 20 minute period of profound anoxia following angiocardiography under anesthesia induced with sterile thiopental sodium U. S. P. At autopsy the heart showed the tetralogy of Fallot. Permission for examination of the brain was denied, but the case is included because of the unequivocal evidence of cerebral injury. In one other of the five cases of the tetralogy observed in the series of 251 autopsies there was a cerebral lesion, i. e., venous thrombosis with encephalomalacia, but in none was there a gastrointestinal ulcer.

The eight cases of congenital anomaly of the central nervous system in this series included two of mongolism, two of meningocele with hydrocephalus, two of caudal

15. Berglund.[1a] Opper and Zimmerman.[7c] Webster.[7d] Hartung and Warkany.[7e]

16. Wenger, F.: Focal Anemia Leukocytosis and Fatty Infiltration of the Liver (So-Called Liver Spots), Arch. Path. **44**:336 (Oct.) 1947.

displacement of the cerebellum, brain stem and spinal cord (Arnold-Chiari syndrome), and one each of cerebral hemiatrophy and microcephaly. Only in the last named was there a gastrointestinal lesion, viz., an acute shallow duodenal ulcer (Case 10). In this instance craniosynostosis was associated with a brain that weighed only 240 gm., compared with the 1,060 gm. weight of the brain of a normal 32 month old child and the 335 gm. weight of that of a newborn. Histologically the granular cell layer of the cerebellum was poorly developed, the Purkinje cells were degenerating, and the normal structure of the cerebral cortex was absent.

Not a single gastroduodenal lesion was found in the six cases of congenital hydrocephalus (Table 3), although two examples of such coincidence are reported in the literature.[17] Nor was a gastric or a duodenal ulcer found in any of the eight cases of encephalopathy with icterus (kernicterus), with or without erythroblastosis.

Somewhat surprising was the absence of any acute peptic ulcers in the three cases of brain tumor. This was not so striking when the tumor was a large papilloma in the lateral ventricle, but the other two cases were both instances of astrocytoma of the brain stem, apparently arising in the pons. Such posterior fossa tumors were regarded by Cushing [6] as peculiarly liable to produce gastric or duodenal ulcers. Masten and Bunts [7a] observed gastromalacia in a 10-year-old boy with a posterior fossa tumor; Grant [7b] encountered a hemorrhagic duodenal ulcer in a 10-year-old white boy with a spongioblastoma of the colliculi, and Opper and Zimmerman [7c] noted an esophageal perforation and gastric erosions in a 10-year-old boy with a craniopharyngioma in the tuber cinereum. Recently Mossberger [1d] reported a perforated ulcer of the duodenum of a 5-day-old infant with a hamartomatous growth in the tuberal region of the hypothalamus.

In none of the 10 cases of coexistent cerebral and gastroduodenal lesions was the injury of the brain limited to the hypothalamus. In all there was a more or less widespread injury, often most manifest in the brain stem. Rarely could any change be demonstrated in the paraventricular and supraoptic nuclei of the hypothalamus. The pituitary gland was normal on gross and on microscopic examination.

COMMENT

In a consideration of the probable pathogenesis of these gastroduodenal ulcers it is well to emphasize that they are acute ulcers and that their relation to the chronic peptic ulcer is not under discussion. This restriction, however, still leaves available the great bulk of experimental work, since nearly all gastrointestinal ulcers produced in animals have been acute. The literature on the experimental production and treatment of peptic ulcers has recently been reviewed by Berg.[18]

There is little room for doubt that the proteolytic activity of the gastric juice is an essential factor in the production of acute peptic ulcers and in the maintenance of the chronic ones.[19] Dragstedt [20] stated that when gastric juice is secreted in excessive amounts the neutralizing mechanisms are overcome and ulcer develops.

17. Berglund.[1a] Langlois.[7f]

18. Berg, M.: Am. J. Digest. Dis. **16**:35, 1949.

19. Schiffrin, M. J., and Ivy, A. C.: Physiology of Gastric Secretion, Particularly as Related to Ulcer Problem, Arch. Surg. **44**:399 (March) 1942.

20. Dragstedt, L. R.: Pathogenesis of Gastroduodenal Ulcer, Arch. Surg. **44**:438 (March) 1942.

Ricketts and co-workers [21] found that in man chronic peptic ulcer occurs only in association with an acid gastric secretion. Achlorhydria lasting longer than three months produces complete healing. Perhaps one factor in the relative infrequency of peptic ulcer during infancy is the low concentration of free acid in the gastric juice. According to Miller,[22] the full-term infant has 27 degrees of free acid on the first day of life; this falls to zero by the eighth day; free acid reappears toward the end of the second week but remains at a low level throughout infancy. The importance of age was also noted by Lillehei and Wangensteen,[23] who found that there is a gradation on the basis of age in susceptibility to histamine-induced ulcer in dogs. The oldest dogs exhibited the highest incidence of gastroduodenal ulcers, whereas pups of 3 to 8 months showed the greatest resistance.

It is now generally accepted that stimulation of the vagus nucleus results in the gastric secretion of hydrochloric acid [24] as well as of pepsin.[25] Hence with the vagus as mediator, cerebral lesions have been linked to peptic ulcers through an effect on the quality and quantity of the gastric secretions. This is an old idea and was clearly stated by Rokitansky [5]: "Perhaps the proximate cause may be looked for in diseased innervation of the stomach, owing to a morbid condition of the vagus, and to extreme acidification of the gastric juice."

The importance of the neutralizing effect of the alkaline pancreatic juice and bile in preventing ulceration of the duodenum is now recognized.[26] It is therefore well to note that stimulation of the vagus produces an increased flow of these secretions,[27] as well as of gastric juice. Today the emphasis is shifting from the acid to the pepsin component of the gastric juice in the production of gastroduodenal ulcers.[28]

In 1925 Mogilnitzky [29] reported foci of degeneration in the supraoptic and paraventricular ganglia of the hypothalamus and in the ganglion nodosum of the vagus in four cases of chronic gastric ulcer. This work was subsequently confirmed and extended by Vonderahe.[30] In 1932 Cushing [6] called attention to the coexistence of peptic ulcer and lesions of the brain stem occurring anywhere from the anterior hypothalamus to the ganglion nodosum. A series of papers by other authors reporting similar cases soon followed,[31] and experimental results confirmed the findings

21. Ricketts, W. E.; Palmer, W. L.; Kirsner, J. B., and Hamann, A.: Ann. Int. Med. **30**: 24, 1949.

22. Miller, R. A.: Arch. Dis. Childhood **16**:22, 1941.

23. Lillehei, C. W., and Wangensteen, O. H.: Proc. Soc. Exper. Biol. & Med. **68**:129, 1948.

24. Janowitz, H. D., and Hollander, F.: Proc. Soc. Exper. Biol. & Med. **76**:49, 1951.

25. Best, C. H., and Taylor, N. B.: The Physiological Basis of Medical Practice, ed. 5, Baltimore, Williams & Wilkins Company, 1950.

26. (*a*) Schiffrin, M. J., and Warren, A. A.: Am. J. Digest. Dis. **9**:205, 1942. (*b*) Poth, E. J.; Manhoff, L. J., Jr., and DeLoach, A. W.: Surgery **24**:62, 1948. (*c*) Tanturi, C. H., and Ivy, A. C.: Am. J. Physiol. **121**:270, 1938. (*d*) Wener, J.; Hoff, H. E., and Simon, M. A.: Gastroenterology **11**:904, 1948.

27. Best and Taylor.[25] Tanturi and Ivy.[26c]

28. Schiffrin and Warren.[26a] Wener and others.[26d]

29. Mogilnitzky, B. N.: Virchows Arch. path. Anat. **257**:109, 1925.

30. Vonderahe, A. R.: Histopathologic Changes in the Nervous System in Cases of Peptic Ulcer, Arch. Neurol. & Psychiat. **41**:871 (May) 1939.

31. Miller, J. K.: Am. J. Surg. **41**:474, 1938. Masten and Bunts.[7a] Grant.[7b] Opper and Zimmerman.[7c]

in man.[32] However, Martin and Schnedorf [33] were unable to obtain gastroduodenal ulceration in monkeys and cats when they produced small localized hypothalamic lesions with the Horsley-Clarke apparatus. In reviewing the experimental work Sheehan [34] concluded that large destructive injuries of the interbrain were most likely to precipitate a gastrointestinal lesion. Wener and Hoff [35] in an extensive survey of the neurohumoral aspects of peptic ulcer formation admit that although the balance of facts indicates that damage of the hypothalamus may produce an ulcer, the mechanism is unknown.

The fact that in experimental animals rather extensive traumatizing operations were carried out on the brain in most of the cases in which peptic ulcers subsequently developed raises the question whether shock might play an important role. Among the first to point this out were Penner and Bernheim,[36] who suggested that in at least seven of the 11 cases reported by Cushing the patient was in shock. The intramuscular injection of histamine in wax is a standard method of producing peptic ulcer in animals.[37] Selye noted that during the alarm reaction to stress there may be a rise in the blood concentration of histamine [38] and that gastroduodenal ulcers are characteristic of the shock phase.[39]

The well known Curling's ulcer of the duodenum following extensive burns may be attributed to the profound shock the patients suffer. In a thorough study of the genesis of gastroduodenal ulcer following burns Friesen [40] found that in dogs given injections of histamine phosphate peptic ulcers developed sooner and more frequently when they had also received an extensive superficial burn. He concluded that ulceration occurred only in those animals in which hemoconcentration and mucosal congestion attended the burn. Hemoconcentration, as emphasized by Moon,[41] is one of the cardinal signs of shock. The possible relationship of extensive burns, adrenal damage, and duodenal ulcer was reported by Harris [42] in a 3½-year-old child and experimentally investigated by McLaughlin.[43] The latter demonstrated that severe burns may produce injury to the adrenals.

The importance of the adrenals in the response to stress is receiving much attention, stimulated by the work of Selye and the recent interest in cortisone and the adrenocorticotropic hormone. However, as early as 1913 Finzi [44] reported on

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32. Watts, J. W., and Fulton, J. F.: Ann. Surg. **101**:363, 1935. Hoff, E. C., and Sheehan, D.: Am. J. Path. **11**:789, 1935. Keller, A. D.: Ulceration in the Digestive Tract of a Dog Following Intracranial Procedures: Preliminary Study, Arch. Path. **21**:127 (Feb.) 1936.

33. Martin, J., and Schnedorf, J. G.: Am. J. Physiol. **122**:81, 1938.

34. Sheehan, D.: The Hypothalamus and Gastro-Intestinal Regulation, in The Hypothalamus and Central Levels of Autonomic Function, Baltimore, Williams & Wilkins Company, 1940.

35. Wener, J., and Hoff, H. E.: Canad. M. A. J. **59**:115, 1948.

36. Penner, A., and Bernheim, A.: Acute Postoperative Esophageal, Gastric and Duodenal Ulcerations: Further Study of Pathological Changes in Shock, Arch. Path. **28**:129 (Aug.) 1939.

37. Varco, R. L.; Code, C. F.; Walpole, S. H., and Wangensteen, O. H.: Am. J. Physiol. **133**:475, 1941.

38. Selye, H.: Ann. Int. Med. **29**:403, 1948.

39. Selye, H.: J. Endocrinol. **6**:117, 1946.

40. Friesen, S. R.: Surgery **28**:123, 1950.

41. Moon, V. H.: Am. J. Path. **24**:235, 1948.

42. Harris, R. I.: Brit. J. Surg. **16**:677, 1929.

43. McLaughlin, C. W.: Curling Ulcer: Study of Intestinal Ulceration Associated with Suprarenal Damage, Arch. Surg. **27**:490 (Sept.) 1933.

44. Finzi, O.: Virchows Arch. path. Anat. **104**:413, 1913.

gastric erosions and ulcers that developed in dogs and rabbits subjected to partial or complete adrenalectomy. Furthermore, he described five cases of gastric or duodenal ulcer in man that were associated with a variety of minor lesions in the adrenals. The experimental results of Finzi have been confirmed by other investigators.[45] In his recent book Selye [46] expressed the belief that either an excessive or a defective production of adrenal corticoid hormones may play a part in the formation of peptic ulcers.

The adrenals in the cases reported here showed no consistent pathological change. The weight in nearly all instances was within normal limits, although in some cases the cortex appeared hyperplastic.

Selye [39] pointed out that the pituitary probably occupies the central position in the defense reactions that are essential for resistance and adaptation to nonspecific damage. He further stated that acute gastric ulcers are a part of the adaptation syndrome. One of the characteristics of this syndrome is the secretion of increased quantities of adrenocorticotropic hormone by the pituitary. Clinical support for the belief that this hormone may be ulcerogenic has been offered by several investigators who noted the occurrence of acute perforating duodenal ulcers or the exacerbation of chronic ulcers in patients on adrenocorticotropic hormone therapy.[47]

A partial explanation of this effect may be found in the observation of Creditor and associates [48] that during the hyperadrenalism induced by such therapy there is inhibition of wound healing in man. Another group of investigators [49] studying the effect of cortisone on wound healing in mice reported a complete suppression of all elements of repair in the treated animals as compared with the control group. In view of the effect of pepsin in the experimental production of ulcers noted above, it may be significant that Spiro, Reifenstein, and Gray [50] have presented evidence indicating that the adrenocorticotropic hormone stimulates gastric gland activity by way of the adrenal to produce an increase in gastric pepsin as well as of urinary uropepsin.

The implication of this work on the adrenocorticotropic hormone with respect to the problem of cerebral lesions associated with peptic ulcer is clarified by the experiments of Hume and Wittenstein.[51] They observed that the normal eosinopenic response of dogs to stressing stimuli is modified or abolished by making small electrolytic lesions in the hypothalamus. From this they concluded that the intact hypothalamus is essential for the normal release of the adrenocorticotropic hormone

45. Mann, F. C.: J. Exper. Med. **23**:203, 1916. Stewart, G. N., and Rogoff, J. M.: Am. J. Physiol. **91**:254, 1929. McLaughlin, C. W.: Ann. Surg. **101**:554, 1935.

46. Selye, H.: The Physiology and Pathology of Exposure to Stress, Montreal, Acta, Inc., 1950.

47. Beck, J. C.; Browne, J. S. L.; Johnson, L. G.; Kennedy, B. J., and MacKenzie, D. W.: Canad. M. A. J. **62**:423, 1950. Habif, D. V.; Hare, C. C., and Glaser, G. H.: Perforated Duodenal Ulcer Associated with Pituitary Adrenocorticotropic Hormone (ACTH) Therapy, J. A. M. A. **144**:996 (Nov. 18) 1950. Smyth, G. A.: Activation of Peptic Ulcer During Pituitary Adrenocorticotropic Hormone Therapy, ibid. **145**:474 (Feb. 17) 1951.

48. Creditor, M. C.; Bevans, M.; Mundy, W. L., and Ragan, C.: Proc. Soc. Exper. Biol. & Med. **74**:245, 1950.

49. Spain, D. M.; Molomut, N., and Haber, A.: Science **112**:335, 1950.

50. Spiro, H. M.; Reifenstein, R. W., and Gray, S. J.: J. Lab. & Clin. Med. **35**:899, 1950.

51. Hume, D. M., and Wittenstein, G. J.: The Relationship of the Hypothalamus to Pituitary-Adrenocortical Function, in Proc. First Clin. ACTH Conf., Philadelphia, The Blakiston Company, 1950.

and the adrenal corticoids in response to stress. They further suggested that the hypothalamus is integrated with the pituitary via a humoral rather than a nervous mechanism. This raises the possibility that the neurosecretory cells described by the Scharrers [52] in the supraoptic and paraventricular nuclei of the hypothalamus may be stimulated to increased activity by lesions in the neighboring brain tissue. The resultant concentration of adrenocorticotropic and corticoid hormones may exert an ulcerogenic effect on the gastric or the duodenal mucosa.

CONCLUSIONS

From the foregoing discussion it is manifest that the causative relationship between cerebral lesions and acute gastroduodenal ulcers are multiple and complex. The integrating factors may be nervous or humoral, and perhaps most often both. The neural mechanism is probably that suggested by Cushing, viz., stimulation of a parasympathetic center in the diencephalon that secondarily activates cranioautonomic nuclei, especially the dorsal nucleus of the vagus. The resultant autonomic imbalance produces local ischemia [53] or marked hyperemia with stasis and increased capillary permeability.[26d] Either of these vascular changes leads to anoxia and tissue damage with decreased resistance to the action of the gastric juice. Besides vasomotor control, the motility of the gastrointestinal tract and the quantity and the quality of the digestive juices are under the direct influence of the hypothalamus.[34] Increase in gastric hydrochloric acid or pepsin and/or a decrease in the neutralizing bile or pancreatic juice may be important in the production of gastroduodenal ulcers.

In conjunction with the well known neural mechanism the more recently recognized humoral responses probably play a part. In all the cases in the series here presented the patient was subjected to subacute or chronic stress. The supraoptic and paraventricular nuclei, which may be most active in stimulating the production of adrenocorticotropic hormone, were intact, as was also the pituitary. The adrenals, on which the adrenocorticotropic hormone exerted its effect, likewise were not the seat of morphologically recognizable damage. It is likely that in these patients large amounts of corticotrophin and corticoid hormones were liberated. This may have led to increased gastric secretion and to impaired healing of the erosions initially produced by the focal vascular changes.

SUMMARY

Among 251 consecutive autopsies on infants and children there were seven cases of acute duodenal ulcer, two of gastromalacia, and one case of multiple acute gastric ulcers. All these conditions were associated with cerebral lesions as follows: tuberculous meningoencephalitis, three cases; bulbar poliomyelitis, two; edema and anoxia, two, and intraventricular hemorrhage, sinus thrombosis, and microcephaly, each one.

These patients manifested the alarm reaction of Selye's general adaptation syndrome. The probable relationship of this and of the altered activity of the autonomic nervous system to the production of the gastroduodenal ulcers is discussed.

All photographs were made by Mr. Joe Humphrey.

52. Scharrer, E., and Scharrer, B.: Secretory Cells Within the Hypothalamus, in The Hypothalamus and Central Levels of Autonomic Function, Baltimore, Williams & Wilkins Company, 1940.

53. Bishton, R. L.: Brit. J. Exper. Path. **31**:316, 1950.

OBSERVATIONS ON A CYTOPLASMIC HEPATIC-CELL PIGMENT IN MAN

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THE PRESENCE of such hepatic-cell pigments as hemosiderin (cytosiderin), melanin, lipofuscin and hemofuscin (cytolipochrome) has been associated with disease of the liver.[1] The present report deals with a cytoplasmic liver-cell pigment which was observed in the course of histochemical studies of the human liver. The pigment appears to be a constituent of normal human liver and decreases in amount in certain diseases of the organ.

METHODS AND MATERIAL

Seventy-five needle-biopsy, 12 surgical-biopsy and 131 postmortem specimens of human liver, obtained from 218 patients, have been examined. The tissues have been fixed in the following solutions: Zenker's solution, with and without 5% acetic acid; 95% ethyl alcohol; Carnoy's solution; 10 per cent formaldehyde solution; Bouin's solution, and acetone. Tissue sections, 5 to 10 μ in thickness, have been prepared from paraffin-embedded and frozen specimens in the usual manner. Various histochemical stains have been applied to these specimens. The amount of pigment present in each section has been graded from 0 to 4+ by three independent observers. When divergent estimates were obtained, the values were averaged. In general, there was close agreement between the estimates of the three observers.

RESULTS

Physical Properties.—The pigment is a golden-brown granular deposit, best observed in cleared, unstained sections. The granules are oval to spherical in shape and measure about 1.5 μ in maximal diameter. They are confined to the cytoplasm of the pericentrally located liver cells (Fig. 1), are less abundant in the midzonal cells, and are absent from the cells about the portal areas. The pigment is scattered throughout the cytoplasm and bears no spatial relationship to the nucleus or to the bile canaliculi (Fig. 2). Occasionally the pigment may be seen in larger aggregates in degenerating liver cells. It is not seen in phagocytic cells, bile-duct epithelium or blood-vessel walls. In postmortem specimens the distribution of the pigment is

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1. (*a*) Sheldon, J. H.: Haemochromatosis, London, Oxford University Press, 1935, Chap. 5. (This reference contains an excellent review of the literature.) (*b*) Gillman, J., and Gillman, T.: Structure of Liver in Pellagra, Arch. Path. **40**:239 (Oct.) 1945.

quite uniform from one hepatic lobule to another. This observation lends validity to the pigment grading of the smaller needle-biopsy specimens.

The pigment has not been found in the livers of the dog, the monkey, the rat, the phalanger, and the marsupial *kappi bara*. Organs other than the liver have not been investigated.

Histochemical Properties.—All studies have been made on tissue sections, since methods have not been developed for extracting the pigment from the tissue for direct chemical analysis. Attention was first drawn to this pigment by its distinct

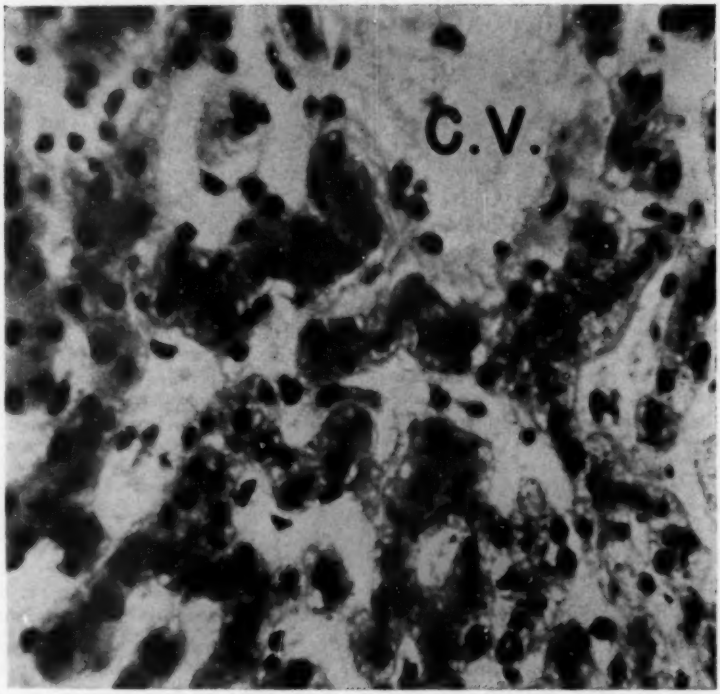

Fig. 1.—The small black cytoplasmic granules in the cells about the central vein (*C. V.*) are pigment bodies stained by the Giemsa dye; microscopic magnification, × 440; photographic enlargement, × 1.6.

basophilism such dyes as azure I, azure II, Giemsa, toluidine blue, and methylene blue. The pigment is stained pink-orange with hematoxylin-eosin and azocarmine but is poorly differentiated by these dyes because of the diffuse cytoplasmic eosinophilia.

Hemosiderin may be confused with the pigment in the unstained section and in sections stained with the basic dyes. However, the two pigments are differentiated by the iron-positive reaction of hemosiderin and the geographic distribution observed within the liver lobule. Hemosiderin is seen in phagocytic cells as well as in liver cells. Furthermore, it is usually deposited in the periportal liver cells, and

as the accumulation increases, it is seen in the pericentral liver cells. To avoid confusion, sections have been stained for iron, without counterstain.[2] Estimates of pigment content have been made from such sections and are reported in Section C.

The pigment is negative to the acid-fast reaction for ceroid.[3] Tests for nucleic acids were negative. The pigment does not absorb ultraviolet rays at 2,600 A., does not stain with the Feulgen technique, is not digested with either ribonuclease or desoxyribonuclease, and does not dissolve in hot trichloroacetic acid.[4] It is negative

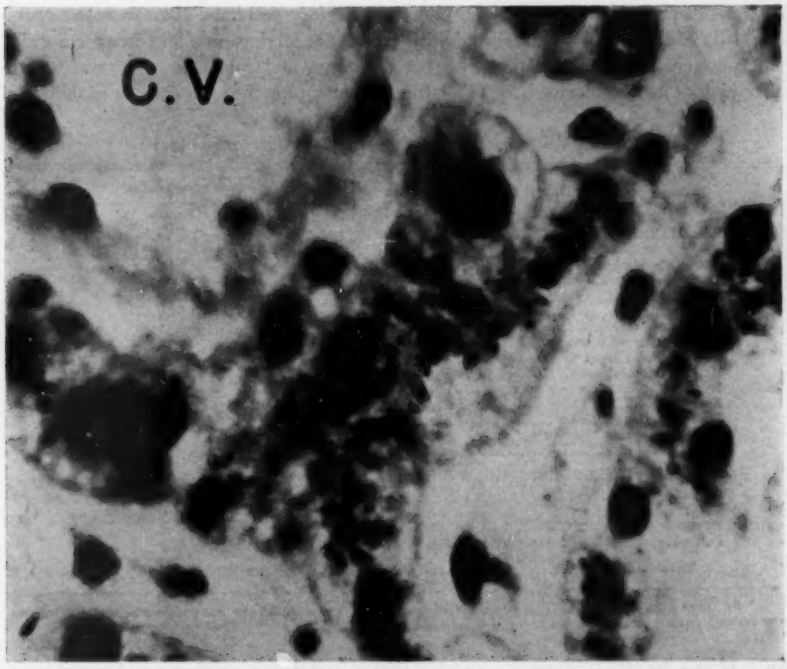

Fig. 2.—The basophilic pigment granules are irregularly scattered throughout the liver cell cytoplasm. This is an oil-immersion photomicrograph of a part of the field shown in Fig. 1 (Giemsa stain); microscopic magnification, × 970; photographic enlargement, × 1.6.

for polysaccharide when stained by the periodic acid method,[5] and it resists digestion with amylase. The pigment is negative to tests for bile[6] and for alkaline phosphatase.[7]

2. Lillie, R. D.: Histopathologic Technic, The Blakiston Company (Division of Doubleday & Company, Inc.), 1948, p. 123.

3. Endicott, K. M., and Lillie, R. D.: Am. J. Path. **20**:149, 1944.

4. Pollister, A. W., and Ris, H.: Nucleoprotein Determination in Cytological Preparations, Cold Spring Harbor, Symp., Quant. Biol. **12**:147, 1947.

5. Glick, D.: Techniques of Histo- and Cytochemistry, New York, Interscience Publishers, Inc., 1949, p. 44.

6. Stern, J.: Compt. rend. Soc. biol. **120**:1136, 1935.

7. Gomori, G.: J. Cell. & Comp. Physiol. **17**:71, 1941.

The pigment is insoluble in petroleum benzin U. S. P., chloroform, dehydrated alcohol N. F., acetone, 5% butyl alcohol and xylene even after 24-hr. extraction. It is insoluble in distilled water, saline solution, and serum at 4 C. and 37 C. up to 48 hr. It resists solution with acid (pH 1.5) and base (pH 10). It is not destroyed in tissue incubated in saline solution and serum at 4 C. and 37 C. up to 48 hr. It resists digestion with trypsin.

When paraffin-embedded or frozen sections are stained with oil red O, heavy pink droplet aggregates obscure the cytoplasmic detail so that a positive pigment reaction cannot be excluded. The pigment does not yield the color reaction characteristic of lipofuscin when treated with potassium ferricyanide and ferric chloride. The pigment is readily stained with basic fuchsin, and subsequent treatment with alcohol does not decolorize it.[8] The latter property plus the pigment's insolubility in

Pigment Distribution and Diagnosis (218 Liver Specimens)

Diagnostic Category	Pigment Distribution			
	0	1-2+	3-4+	Total
Normal liver	7	15	49	71
Fatty liver	11	18	10	39
Laennec's cirrhosis	49	5	0	54
Laennec's cirrhosis with hemochromatosis	5	0	0	5
Biliary cirrhosis	1	0	0	1
Cholangitis	2	0	0	2
Cholecystitis	0	0	1	1
Cholecystitis with stones	0	2	0	2
Carcinoma of head of pancreas	1	0	0	1
Metastatic carcinoma	3	2	0	5
Lymphoma	2	3	1	6
Amyloidosis	0	1	0	1
Lupus erythematosus *	2	0	0	2
Chronic passive congestion *	1	1	14	16
Premature infant *	1	0	0	1
7-week infant *	1	0	0	1
Tuberculosis *	0	2	0	2
Hemangioma *	0	1	0	1
Focal necrosis *	0	1	0	1
Hepatitis *	0	2	2	4
Periportal lymphocytic infiltration *	0	0	2	2
Total	86	53	79	218

* All these data are derived from postmortem specimens. The data of the remaining diagnostic categories are derived from postmortem and biopsy specimens.

fat solvents and its basophilism suggest that it is identical with the pigment hemofuscin. However, hemofuscin is reported soluble in 5% hydrogen dioxide,[8] while the pigment under discussion is insoluble in H_2O_2.

Neither the method of fixation nor that of embedding (paraffin or frozen blocks) alters the appearance or the amount of pigment in any particular specimen. Lastly, the pigment appears the same in liver tissue obtained in needle biopsy, surgical biopsy, and postmortem examination.

Relation to Disease of the Liver.—In the course of this study it was found that the amount of pigment varied from one specimen of liver to another. Accordingly, the distribution of the pigment was studied in 87 biopsy and 131 postmortem liver specimens. Estimates of pigment content were graded from 0 to 4 +. For purposes of presentation, specimens have been grouped in three categories: 0, 1 + and 2 +, 3 + and 4 +, and the biopsy and postmortem specimens have been tabulated together (table).

8. Mallory, F. B.: Pathologic Technic, Philadelphia, W. B. Saunders Company, 1938, p. 136.

Of 71 normal specimens, about 90 per cent had pigment. Seventy per cent had pigment in abundance (3 + and 4 +). Only 10 per cent had no pigment. Of the fatty livers, about 30 per cent had no pigment, and about 30 per cent had pigment in great abundance (3 + and 4 +). However, of the cirrhotic livers, only about 10 per cent had pigment, and about 90 per cent were pigment-free (Fig. 3). The data on the remaining miscellaneous specimens are presented for completeness, but it is difficult to draw conclusions since the numbers of cases in the various groups are so small (table). It is of interest that in livers with chronic passive congestion abundant pigment was present. In four cases of severe hepatitis pigment was present.

All specimens were from adults in their third to ninth decades, except for two, which were from a premature infant and a 7-week-old infant. These two specimens

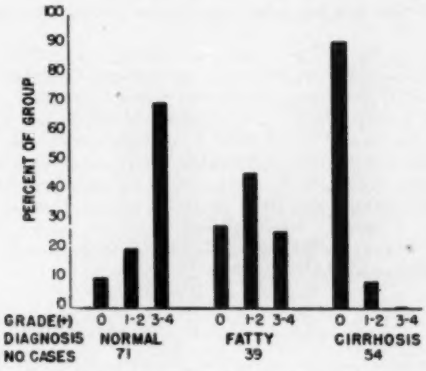

Fig. 3.—The frequency distribution of pigment estimates in normal, fatty, and cirrhotic livers.

contained none of the pigment under discussion, although hemosiderin was present in abundance. In the other specimens the distribution of pigment was not related to the ages of the patients.

Studies have been made of the pigment distribution in 13 alcoholic patients on whom biopsy was performed two to six times during periods of one to 14 mo. Initially the pigment content was reduced in most of these specimens, especially when fatty infiltration was present. However, it was noted that even though the fat might clear in later biopsy specimens, the amount of pigment did not increase. These limited observations suggest that the loss of pigment is an irreversible phenomenon.

COMMENT

The various pigments which have been described in the liver have been associated with disease processes. Thus the pigments hemosiderin (cytosiderin), lipofuscin, and hemofuscin (cytolipochrome) have been associated with disturbances in iron metabolism,[1a] "wear and tear processes," [1a] and the pathogenesis of a type of cirrhosis and hemochromatosis,[1a, b] respectively. The pigment under discussion differs from the latter group of pigments in its general histochemical properties, although in many respects it is similar to hemofuscin. However, it belongs to a different class of pig-

ments in that the data show that about 90 per cent of normal livers contain the pigment. It is seen in decreasing amounts in certain diseases of the liver. In the fatty livers and especially in those livers with Laennec's cirrhosis the pigment may be reduced or absent. With the clearing of fat the amount of pigment does not increase. In other disease states alterations of pigment content may be seen. These data suggest that the pigment is a metabolically active complex in normal human-liver cells. Its chemical identity is unknown. Studies are in progress on methods of extracting the pigment for direct chemical analysis.

SUMMARY

A cytoplasmic pigment of the human-liver cell has been observed as a constituent of normal liver tissue. It disappears from fatty and cirrhotic livers.

Some of its histochemical properties have been described.

It is suggested that this pigment is an active complex in the metabolism of the liver cell.

Prof. A. W. Pollister of the department of zoology, Columbia University, aided this investigation with ultraviolet spectrophotometric studies. Dr. Paul Klemperer, director of pathology, Mt. Sinai Hospital, New York, contributed criticisms and suggestions, and supplied some of the tissue specimens. Dr. William A. Antopol, director of pathology, Beth-Israel Hospital, New York, and Dr. Ted Ehrenreich of the department of pathology, Veterans Administration Hospital, Bronx, N. Y., made some of the tissue specimens available. Dr. Ross Negrelli, director of pathology, New York Zoological Society, made some of the animal specimens available. Dr. J. Murray Steele, director, Research Service, New York University, Goldwater Memorial Hospital, made suggestions relating to the pursuit of those studies and to the preparation of the manuscript.

A METHOD FOR THE HISTOPATHOLOGIC STUDY OF THE ATRIO-VENTRICULAR NODE, BUNDLE, AND BRANCHES

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THERE is at present no routine method for the histologic study of the human atrioventricular node, bundle, and bundle branches. Either serial sections are employed, or scattered, helter-skelter sections are made. The first method is not feasible for routine work, while the second is erratic.

In the last two years we have been engaged in devising a method which would give 100% continuity of structures and at least 90% yield of pathologic changes. Toward that end we have presented the gross and histologic structure of these parts in various age groups.[1] The present report deals with a qualitative method of studying these structures. The quantitative analysis of this method is now being developed and will be forthcoming later.

MATERIALS AND METHODS

The exact course of the a-v node, bundle, and bundle branches was ascertained by gross dissection. By trial and error, various angles of cutting were used, and finally the one described below was adopted. By this method, 55 normal hearts were cut and studied histologically. It was found that 100% continuity of parts could be obtained in these hearts by this method.

The heart is opened at autopsy in the conventional method. The heart, its chambers packed with cotton, is fixed for 6-24 hr. in 4% formaldehyde solution. The parietal and inferior walls of both atriums and ventricles are then cut away, leaving the septal wall of the chambers. The pars membranacea is observed carefully, as well as the Eustachian valve, the coronary sinus, the central fibrous body, the muscle of Lancisi and the crista supraventricularis. This gives the general location and direction of the atrioventricular node, bundle, and right branch. The base line for cutting, Cut 1 (Fig. 1*a*), is made along a line about ¼ to ½ in. (6 to 12 mm.) below the moderator band to produce an angle of about 45 degrees with the line of the crista. Cut 2 is made at right angles to Cut 1 and passes through the junction of the Eustachian valve and the limbus. Cut 3 is made at right angles to the second cut, passing through the base of the aorta. Cut 4 is made just lateral to the moderator band at right angles to the third cut. This leaves a rectangle of muscle containing all of the desired structures with the exception of the lower aspect of the left bundle-branch fasciculi (Fig. 1*b*).

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This investigation was supported by a research grant (H-430C) from the National Heart Institute of the National Institutes of Health, United States Public Health Service.

1. (*a*) Widran, J., and Lev, M.: The Dissection of the Atrio-Ventricular Node, Bundle, and Bundle Branches in the Human Heart, to be published. (*b*) Erickson, E. E., and Lev, M.: Aging Changes in the Human Atrio-Ventricular Node, Bundle, and Bundle Branches, to be published.

Cut 5 is made at right angles to the base line, passing through the middle of the pars membranacea. This yields Block 1. Cut 6 is made at right angles to the base line, passing through the muscle of Lancisi. This yields Block 2. Cut 7 is made at right angles to the base line, dividing the remainder of the tissue into Blocks 3 and 4. Tissue sectioning is done at 5 μ thickness. Every 20th section is saved through Block 1, and every 40th section thereafter. These sections are stained with hematoxylin and eosin. No other stains are necessary for identification. Elastic-Van Gieson and reticulum stains may be used as aids in the identification of the right branch. This method yields from 150 to 200 sections of the adult heart.

IDENTIFICATION OF STRUCTURES

All structures are identified by position and the following general characteristics: (*a*) sheath, (*b*) cytoplasm of cells, and (*c*) elastic network. In addition, some parts have further differentiating characteristics.

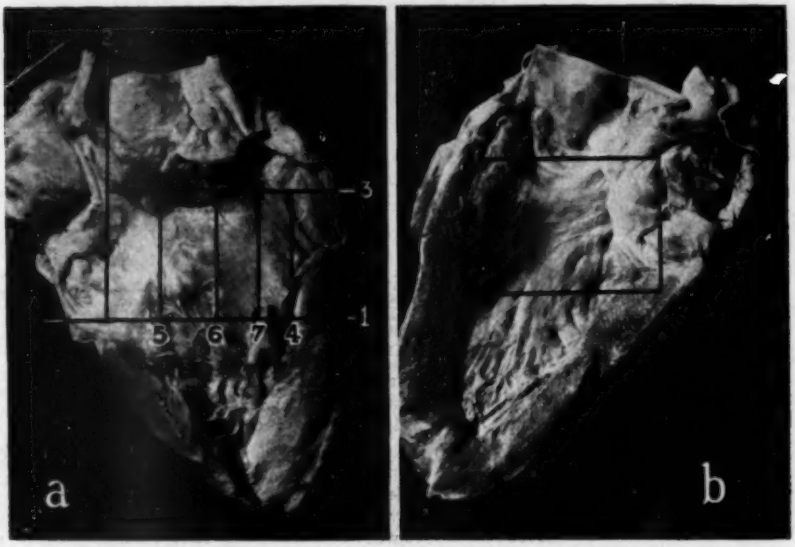

Fig. 1.—Method of sectioning for histopathologic study of the atrioventricular node, bundle and bundle branches: (*a*) Right ventricular and atrial view. (*b*) Left ventricular and atrial view. Numbers *1* to *7* are the cuts referred to in the text.

Sheath.—All parts contain endothelium-lined structures which surround and interdigitate between the fasciculi (Fig. 2*a*). This gives all structures an increased cellularity, which is most marked in the atrioventricular node and bundle. From the end of the node to the termination of the bundle branches, the sheath surrounds a distinct lobulated space (Fig. 2*b*).

Cytoplasm of Cells.—The cytoplasm of the muscle fibers of all parts is less eosinophilic.

Elastic Network.—At any age level the entire system shows an increase in elastic fibers as compared with the musculature of the atriums and the ventricles (Fig. 3). This feature does not have to be relied on for identification. The distal part of the right bundle branch, however, may be more easily identified by this means.

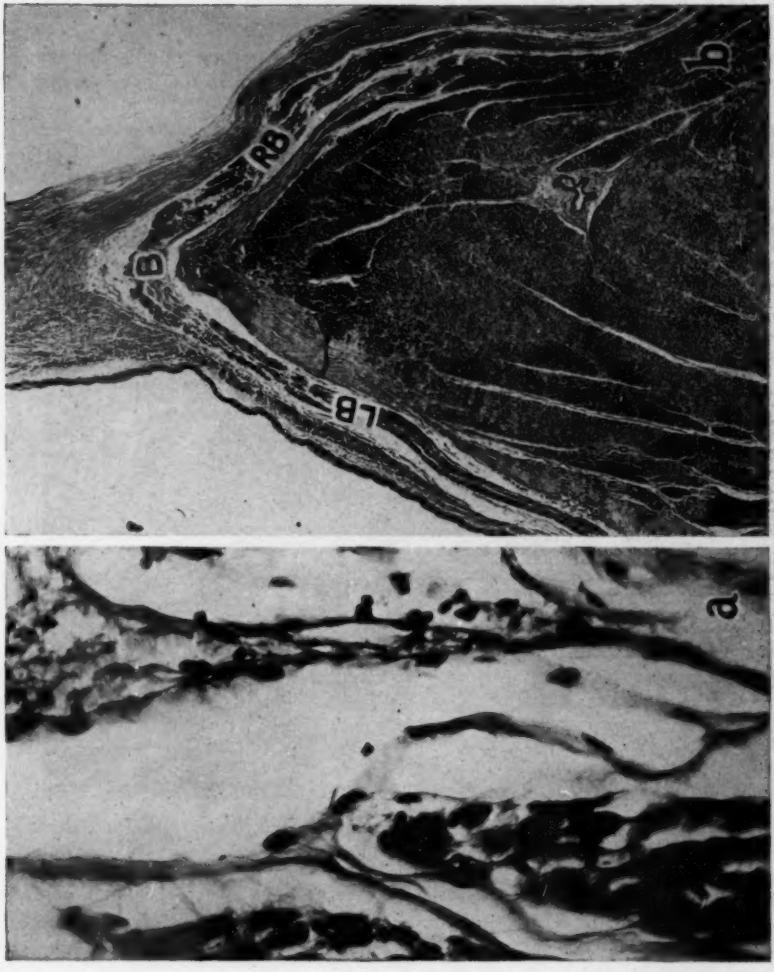

Fig. 2.—Photomicrographs showing the sheath and enclosed space around the bundle and bundle branches: (*a*) Sheath; hematoxylin-eosin stain; × 900. (*b*) Bundle and right and left bundle branches; hematoxylin-eosin stain; × 20. *B* indicates bundle; *RB*, right bundle branch; *LB*, left bundle branch.

FURTHER INDIVIDUAL IDENTIFYING CHARACTERISTICS

Node.—This lies adjacent to the central fibrous body above the base of the tricuspid valve, on the right side of the ventricular septum (Fig. 4). In addition to presenting the above general characteristics, it consists of a loose network of small fibers, which are smaller than the atrial and ventricular fibers postnatally. With the above manner of cutting, it is seen in oblique section.

Bundle.—This consists of two parts: the penetrating portion passing through the central fibrous body (Fig. 5*a*) and the branching portion in the lower part of

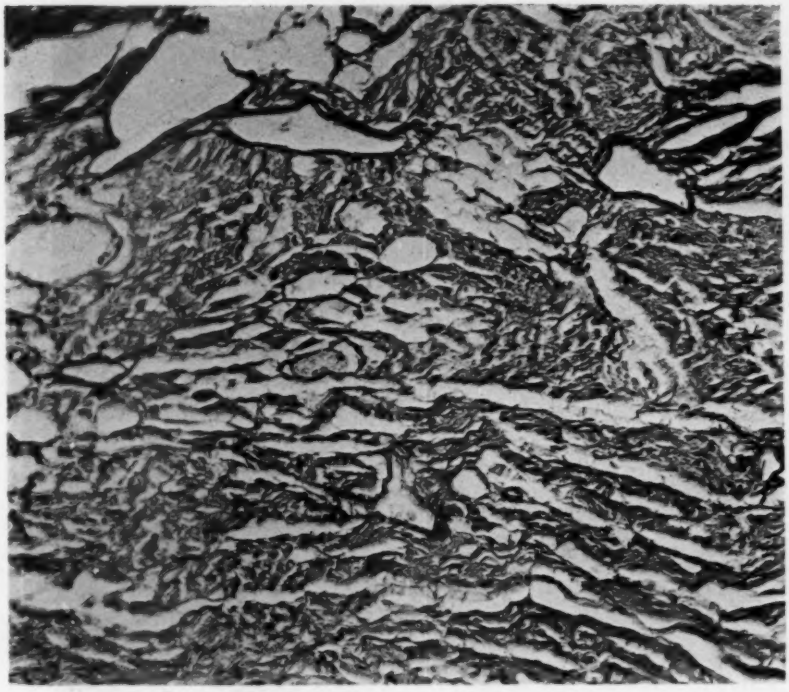

Fig. 3.—Photomicrograph of node, showing increase in elastic tissue. Weigert's elastica with Van Gieson's connective-tissue stain; × 180.

the pars membranacea above the ventricular septum (Fig. 5*b*). The latter part may, however, lie on the left side of the septum and, uncommonly, on the right. Its fibers are likewise smaller than those of the atriums and the ventricles. With the above-described manner of cutting, it is seen in cross section.

Left Bundle Branch.—The fibers are seen on the left side of the septum beneath the endocardium. These fibers are divided into anterior and posterior radiations. With the above manner of cutting, the proximal fibers are seen in oblique section (Fig. 6), while the distal fibers are seen in longitudinal section (Fig. 7). The fibers become progressively larger as they proceed apexward. The larger fibers are

larger than those of the left ventricular myocardium. Further differentiating features, unnecessary in routine work, are the thicker reticular and glycoprotein components of the basement membranes of the fibers.

Right Bundle Branch.—The identification of this branch, in addition to the above characteristics, is dependent on the follow-up of its course in consecutive

Fig. 4.—Photomicrograph of node. Hematoxylin-eosin stain; × 20. *N* indicates node; *A*, atrial musculature; *V*, ventricular musculature.

sections. Its first part is usually subendocardial and easily recognized on the right side of the ventricular septum (Fig. 8*a*). Its fibers are slightly smaller than, or equal to, those of the adjacent myocardium. As it becomes intramyocardial, it can be recognized by the difference in the direction of its fibers as compared with the

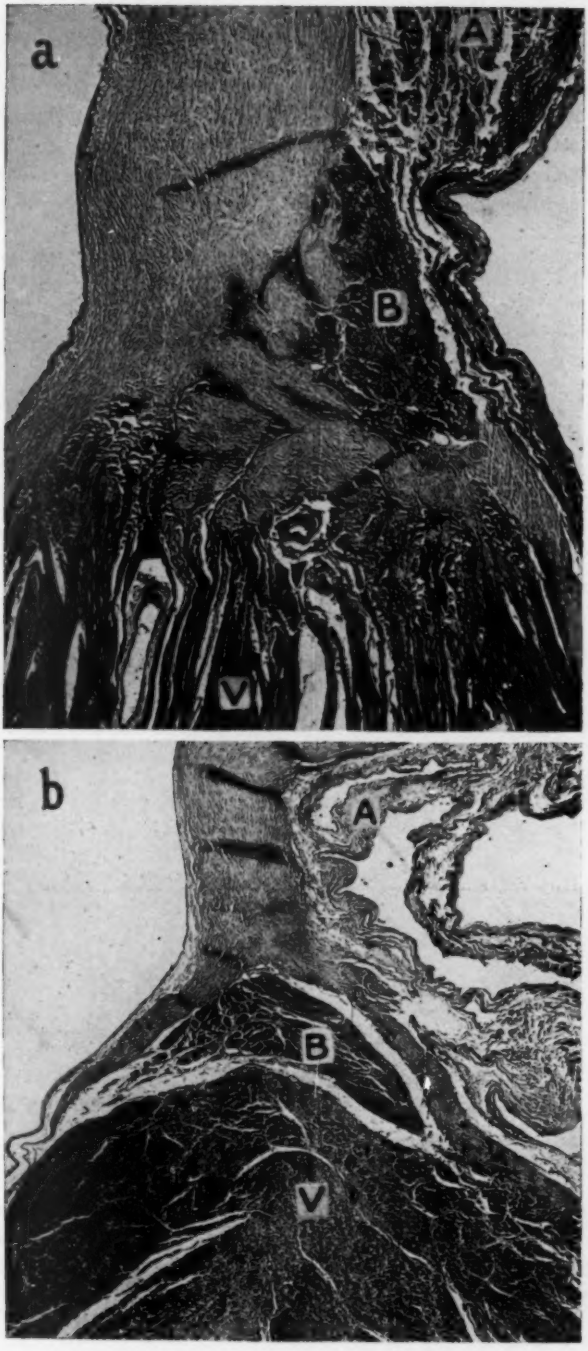

Fig. 5.—Photomicrographs of bundle: (*a*) Penetrating portion; hematoxylin-eosin stain; × 50. (*b*) Branching portion; hematoxylin-eosin stain; × 20. *B* indicates bundle; *A*, atrial musculature; *V*, ventricular musculature.

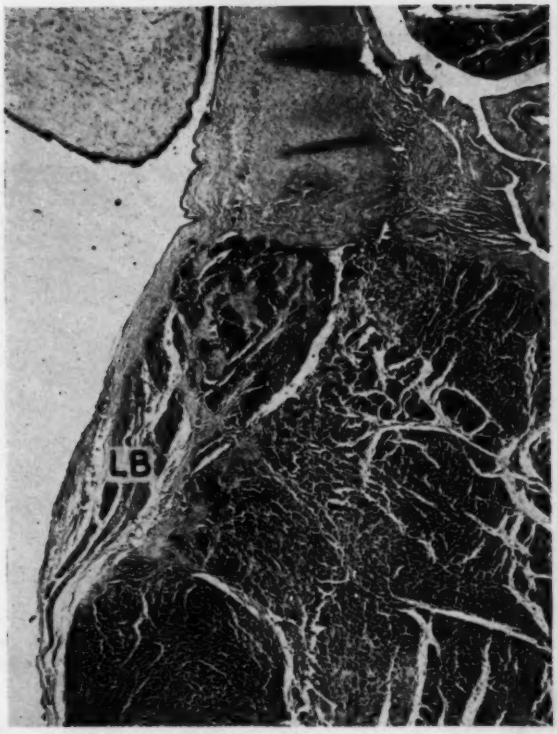

Fig. 6.—Photomicrograph of left bundle branch taken at its origin from bundle. *LB* indicates left bundle branch. Hematoxylin-eosin stain; × 20.

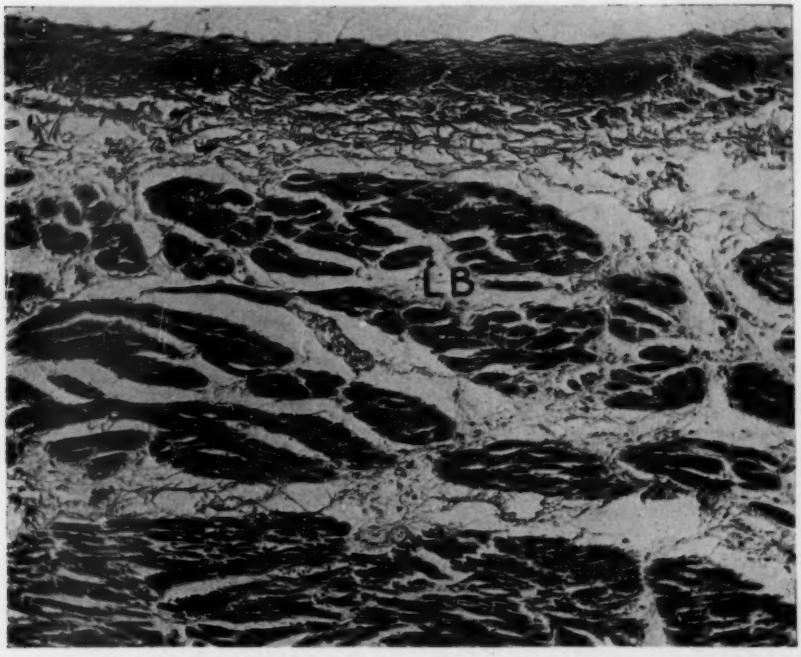

Fig. 7.—Photomicrograph of left bundle branch: lower portion. Hematoxylin-eosin stain; × 100. *LB* indicates left bundle branch.

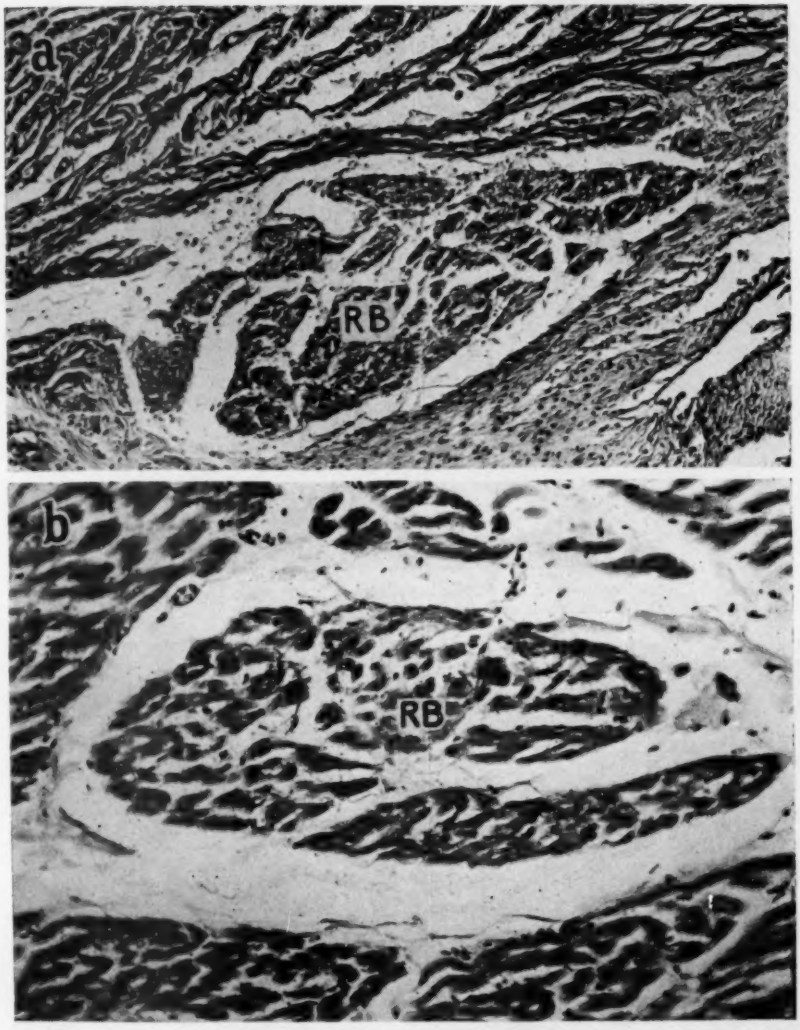

Fig. 8.—Photomicrographs of right bundle branch: (*a*) Proximal portion (newborn); hematoxylin-eosin stain; × 100. (*b*) Distal portion (adult); hematoxylin-eosin stain; × 200. *RB* indicates right bundle branch.

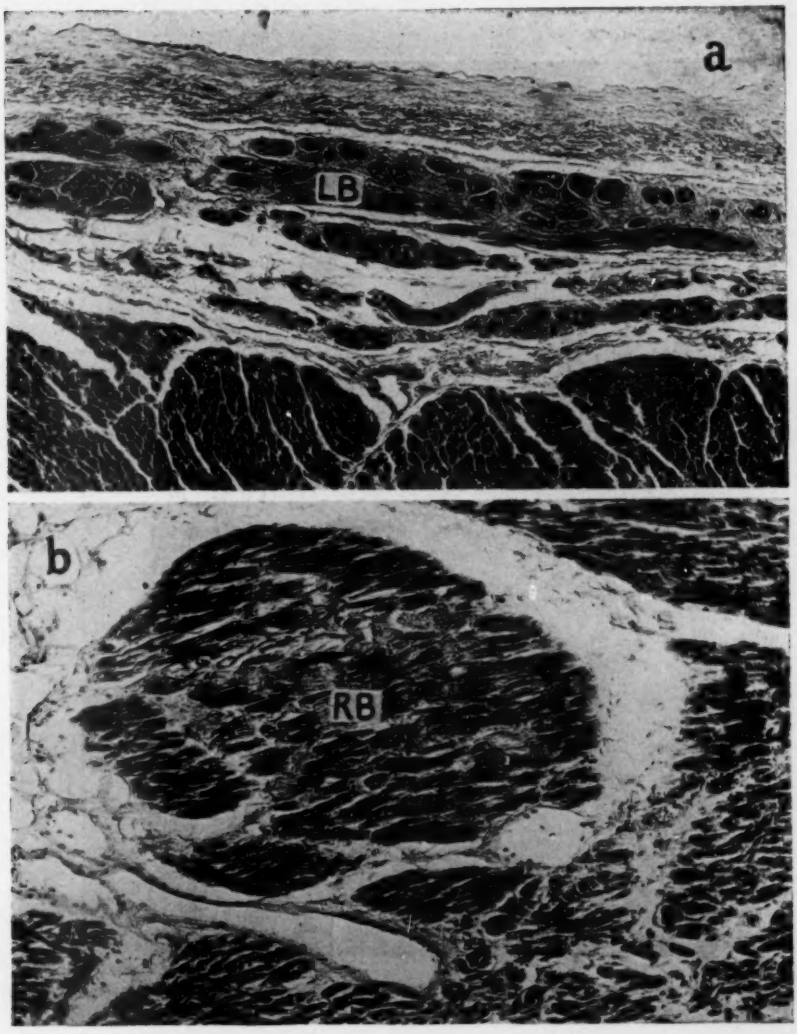

Fig. 9.—Photomicrographs showing an increase in connective tissue: (*a*) Left bundle branch; hematoxylin-eosin stain; × 100. (*b*) Right bundle branch; hematoxylin-eosin stain; × 200. *RB* indicates right bundle branch; *LB*, left bundle branch.

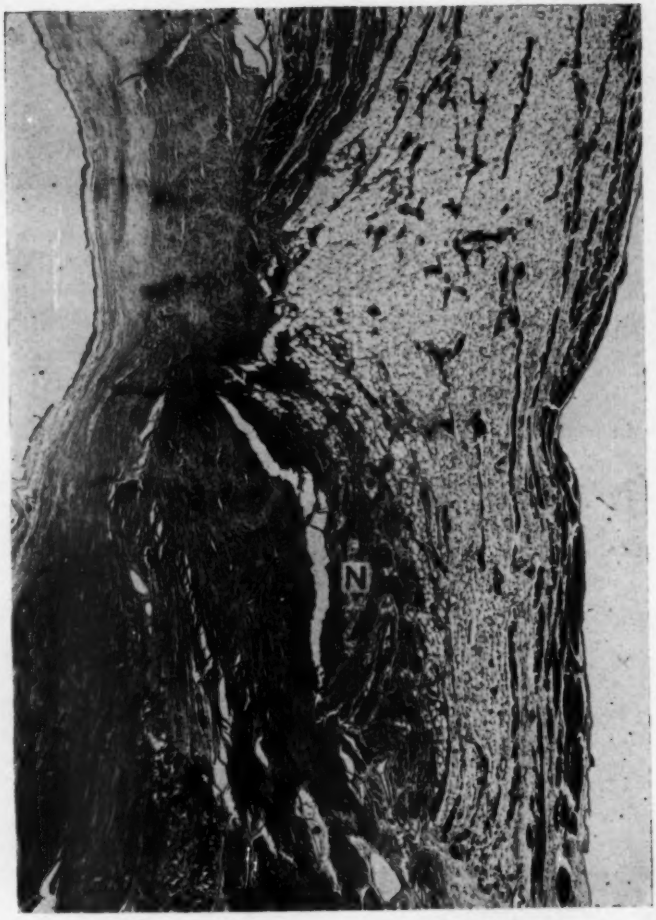

Fig. 10.—Photomicrograph of node, showing an infiltration of fat. Hematoxylin-eosin stain; × 20. *N* indicates node.

surrounding fibers (Fig. 8*b*). A reticulum stain may be used to show this direction more clearly. In its third portion it becomes subendocardial again and its fibers are larger and begin to resemble Purkinje fibers.

NORMAL AGING CHANGES

It is important to differentiate "normal" aging changes from pathologic change. The former have been reported elsewhere.[1b] They are (1) an increase of connective tissue (Fig. 9), (2) an increase of elastic tissue, (3) an infiltration of fat, and (4) an increase in density of reticulum. Fat is found after 30 yr. of age and is usually limited to the node, the bundle, and the beginning of the left and right branches (Fig. 10). The other findings are also found in the remainder of the branches.

SUMMARY

A routine qualitative method is described for the study of the atrioventricular node, bundle and branches.

Criteria are presented for the identification of the various parts.

Mrs. Helen Klassen gave technical assistance in this work.

GRANULOMATOUS GLOMERULONEPHRITIS ASSOCIATED WITH POLYARTHRITIS

Report of a Case

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DURHAM, N. C.

A CASE of a peculiar chronic glomerulonephritis is described in this report. The late stages of glomerular disappearance were associated with the formation of granulomas. Clinically, the patient showed chronic polyarthritis and mild eosinophilia. At autopsy, besides the unusual glomerulonephritis, there was acute necrotizing arteritis, observed in the urinary bladder, the spleen, the ovary and the small intestine, as well as a peculiar deformity of the aortic valve.

Polyarthritis dominated the early clinical picture. Daugherty and Baggenstoss [1] have lately collected seven cases of subacute and chronic glomerulonephritis associated with rheumatoid polyarthritis. Their autopsy studies describe no renal lesions similar to these reported here. Craciun [2] has indicated the association of nephritis and rheumatism. As pointed out elsewhere,[3] Craciun's cases are more like pyelonephritis, possibly following the so-called benign interstitial nephritis.

Daugherty and Baggenstoss have recalled that Osler [4] in 1904 described the kidneys as being involved in the visceral manifestations of the erythema group of skin diseases. Tremaine [5] in 1934 reported a case of polyserositis with arthritis and glomerulonephritis. The association of these types of lesions has suggested to Daugherty and Baggenstoss that their cases be considered as examples of "collagen disease," several features of which appear in the present case.

CLINICAL HISTORY

A white woman, aged 52, was first seen in August, 1947, complaining chiefly of pain, heat, and swelling of knee, ankle, and phalangeal joints of two months' duration. She then received fairly close observation through 11 hospital admissions until her death, in May, 1950, almost three years later.

From the Department of Pathology, Medical College of Alabama, Birmingham.

1. Daugherty, G. W., and Baggenstoss, A. H.: Syndrome Characterized by Glomerulonephritis and Arthritis, Arch. Int. Med. **85**:900-923 (June) 1950.

2. Craciun, E. C., and others: Localisation rénale de la maladie de Bouillard, Ann. anat. path. **10**:363, 1933.

3. McManus, J. F. A.: Medical Diseases of the Kidney, Philadelphia, Lea & Febiger, 1950.

4. Osler, W.: On the Visceral Manifestations of the Erythema Group of Skin Diseases, Am. J. M. Sc. **127**:1-23, 1904.

5. Tremaine, M. J.: Subacute Pick's Disease (Polyserositis) with Polyarthritis and Glomerulonephritis, New England J. Med. **211**:745-749, 1934.

The patient was said to be in her usual fair state of health until April, 1947. In addition to the symptoms of polyarthritis, chills, epistaxis, sinus pain and earache were present.

The past medical history contains a description of "growing pains" at the age of 10 years, with sequelae. For several years prior to 1947 the patient had episodes of tachycardia. The menopause preceded the present illness by six years. A history of contact with raw milk had been elicited. Agglutination tests, including those for typhoid (Salmonella typhosus), paratyphoid (Salmonella schottmuelleri and S. choleraesuis), Proteus OX19 and Brucella antigen, were performed before admission. No significant positive agglutinations were recorded. A pulmonary apical shadow had been observed on a roentgenogram six weeks prior to admission. This had resolved.

The patient had been pregnant four times and had given birth to three children. One son had pulmonary tuberculosis, inactive at present. The husband is living and well. Both parents died of pulmonary tuberculosis. The patient had always been a housewife, doing her own work.

Physical examination revealed a 96 lb. (43.5 kg.), diminutive, fairly well-nourished woman with a normal temperature. The positive findings were limited to slightly swollen, painful, and erythematous proximal phalangeal joints.

The hospital course was uneventful. Therapy consisted in the administration of acetylsalicylic acid, vitamins, etc. No elevation of temperature was recorded. The patient did not receive sulfonamide compounds during the course of this disease.

Laboratory examination revealed an elevated corrected erythrocyte sedimentation rate of 48 mm. per hour, 4,310,000 red blood cells, a hemoglobin content of 75%, a color index of 0.87, and 9,000 white blood cells, with a normal differential count. A complete urinalysis gave negative results. Heterophil antibody was not demonstrated. Blood cultures were repeatedly negative.

Subsequent to discharge the patient experienced a continuation of the polyarthritis and nasal sinus pain. A submucous resection was performed in October, 1947. As an infection developed at the site of operation, the patient was admitted again in November, 1947, for penicillin control of this infection. Unusual findings pertaining to the general systemic condition were limited to evidence of secondary anemia. The red-blood-cell count was 3,610,000, with the hemoglobin content 68% and the color index 0.94. Seven eosinophilic granulocytes were present in the differential count.

The fourth and fifth admissions, January and April, 1948, were marked by fever, general malaise, and cough. An area of patchy pulmonary infiltration was observed in the left lung during the January admission. A diagnosis of atypical pneumonia was rendered. Polyarthritis persisted. Therapy included the administration of streptomycin and penicillin. The erythrocyte-sedimentation rate was 44 and 48 mm. per hour. Eosinophil counts varied between 6 and 12 per cent in four separate blood-cell studies. Blood cultures, sputum examinations, urinalysis and agglutination tests gave essentially negative results. Total serum protein was 6.3 gm., albumin 3.3 gm. and globulin 3.0 gm. per 100 cc.

The sixth through the ninth hospital admissions, occurring during the last four months of 1949, were concerned with the development of bilateral corneal ulcers, and their unsuccessful attempted repair. During these admissions there was continued evidence, both subjective and objective, of polyarthritis. Laboratory examination revealed during this time an eosinophilia of 5 to 9%. The recorded red-blood-cell counts did not fall below 4,300,000 during this period; however, the patient was receiving blood transfusions periodically. The urine remained negative for traces of albumin.

The terminal two admissions, December, 1949, and May, 1950, were of short duration, and observations were not complete; however, anemia and renal failure were becoming obvious. The patient died within a few hours of the last admission, complaining of severe low back pain. The urine revealed albumin (4+) and whole blood. The red-blood-cell count was 2,880,000, with the hemoglobin content 48%.

AUTOPSY (A. H.)

Gross Findings.—At autopsy the skin and the mucous membranes were remarkable only for a generalized pallor. No subcutaneous nodules were observed. External examination revealed extensive bilateral corneal ulcerations and a post-

operative depression defect of the bridge of the nose. Teeth were absent. The proximal phalangeal joints appeared slightly prominent in both upper and lower extremities. The remainder of the surface examination showed essentially normal conditions except that the skin and subcutaneous tissues were friable.

The peritoneal cavity contained approximately 200 cc. of clear fluid. The left pleural space contained 600 cc. and the right pleural space 300 cc. of gray watery fluid. The pericardial space contained no excess fluid. Minor fibrous pleural adhesions were encountered at the left and the right pleural space base and at the apex of the left lung. A moderate fibrinous roughening was observed on all pericardial surfaces.

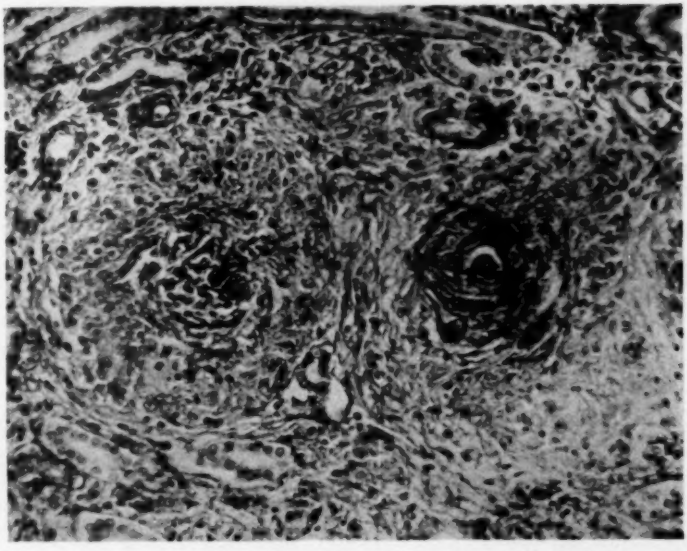

Fig. 1.—Two glomeruli showing different stages of development of granuloma. That at the left is taken to be more advanced. Periodic acid-Schiff stain with hematoxylin counterstain.

The heart weighed 420 gm. and showed hypertrophy of the left ventricle. All valves except the aortic valve and all endocardial surfaces except those in the region of the aortic valve appeared normal. The aortic-valve cusps were all shortened without being greatly thickened. The commissures between posterior and right, and right and left cusps were partially fused. The opening of the right coronary artery was of pinpoint size, but the coronary vessels were patent throughout.

Except for patchy pleural thickening the lungs were not remarkable. The liver was moderately congested. The kidneys were symmetrically reduced in size and had pale cortices. Melanosis of the cecum and moderate sclerosis of the aorta were present.

Microscopic Observations.—The chief microscopic findings of interest are those in the kidney. (See Figs. 1 and 2.) The renal lesions are not those which we

have been accustomed to find with subacute or chronic glomerulonephritis. There is no pseudotubule formation. The chief peculiarity lies in the obsolete and obsolescent glomerulus forming the center of an epithelioid reaction. This structure is more like a tubercle or the subcutaneous nodule of rheumatoid arthritis than it is like the usual obsolete or obsolescent glomerulus in glomerulonephritis. In the particular type of glomerular obsolescence in this case we have been unable to find any definite arterial lesion. There is the impression that the afferent arterioles may be furnishing the nidus for the whole reaction. In this circumstance the case suggests one of periarteritis nodosa or a case of one of the related collagen diseases.

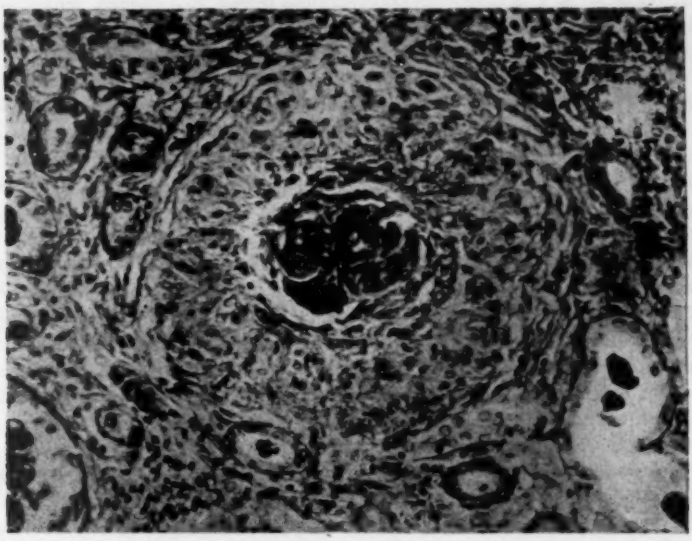

Fig. 2.—Fully developed granulomatous reaction about an obsolete glomerulus. Periodic acid-Schiff stain with hematoxylin counterstain.

The lesion of acute active periarteritis nodosa was present in vessels within the muscularis of the urinary bladder, the spleen, the ovarian stroma, and the serosa of the small bowel. In all these sites polymorphonuclear granulocytes were noted invading and surrounding a partially necrotic arteriolar wall.

The microscopic section of the lesion of the aortic valve shows a heavy round-cell and an occasional eosinophil invasion of the base of the valve. Fibrous proliferation and patchy destruction of elastic-tissue fibrils were also present. Few scattered myocardial polymorphonuclear granulocyte foci were observed, apparently unrelated to vessels. A fibrinous reaction incorporating polymorphonuclear cells was present on the epicardial surface.

Fibrous-tissue proliferation was seen in the right pulmonary apex. The liver was congested, with mild evidence of central necrosis.

There was no gross or microscopic evidence of tuberculosis.

COMMENT

The most striking feature of the case is the peculiar glomerular lesion. The possibility that this case is related to the group of "collagen diseases" is next in importance. These points of interest will be discussed in this order.

The glomerular lesion is striking and in our experience unique. The disappearance of a glomerulus seems rather protean in its early stages. A few pseudotubular structures in a crescent can be recognized in one glomerulus. This is the only feature resembling the usual glomerular obsolescence in subacute or chronic glomerulonephritis which was found in very many sections of these kidneys. Within a little while the glomerulus, already nearly obsolete, and showing in the hematoxylin-eosin section as a hyaline knot, becomes the center of a chronic inflammation, distinguished first by lymphocytes with a few macrophages (Fig. 1, right). The macrophages increase in number and soon arrange themselves in a pattern much resembling that of a hard tubercle. The center of the nodule is formed by the remnants of the glomerulus, now considerably reduced in quantity (Fig. 1, left). All stages of glomerular disappearance and many tuberclelike structures can be found in section. In each well developed tubercle the remnants of a glomerulus occupy the center (Fig. 1, left; Fig. 2). No giant cells are found. There is no collar of lymphocytes around the glomerular lesion at any stage. It appears to be an expanding lesion compressing adjacent tubules (Figs. 1 and 2).

The tuberclelike lesions differ in certain respects from those of Mycobacterium tuberculosis—no caseation and no giant cells, and no tubercle organisms on acid-fast stains. The latter detail is recognized to be nonconclusive, but there is the additional feature of no tuberculosis elsewhere in the body. A tuberculous infection restricted to the glomeruli would be difficult to imagine. When a glomerulus is affected in miliary hematogenous tuberculosis, the lesion is quite different from that present in this case, being characterized by glomerular-loop thrombosis and necrosis, slight caseation, giant cell formation, etc.

This lesion might be classed by some as sarcoidosis. It is felt that grouping nontuberculous tubercles as sarcoid does not advance knowledge much. There is no other evidence of sarcoidosis and none of syphilis. In generalized sarcoidosis, glomerular involvement may take a considerably different form. No organisms or fungi are present in the glomeruli at any stage.

The absence of the usual features of subacute or chronic glomerulonephritis has already been mentioned in passing. As Harvey and MacCallum have described, and as has recently been illustrated adequately,[2] the usual course of glomerular obsolescence in subacute and chronic glomerulonephritis is marked by pseudotubular formations in the epithelial "crescent." In only one glomerulus was this found in the present case in many sections which were studied. The case under discussion is unlike the usual case of glomerulonephritis in the absence of pseudotubule formation as well as in the presence of the granulomatous glomerular lesions.

The case clinically and at autopsy showed several of the features of the "collagen diseases." This term was introduced by Klemperer[6] to characterize a variety of diseases, including lupus erythematosus disseminatus, polyarteritis nodosa, rheumatic fever and rheumatoid arthritis, and others. As Klemperer

6. Klemperer, P.: The Concept of Collagen Disease, Am. J. Path. **26**:505-519, 1950.

pointed out, "All we wanted to express originally was that in certain diseases anatomical investigations reveal conspicuous alterations of the intermediary substances of the connective tissue in a systemic manner."

The peculiar lesion of the aortic valve might fit in with a healed stage of the atypical endocarditis of Libman and Sacks.[7] The aortic cusps were shortened without the thickening characteristic of syphilis. There was fusion of cusps in two of the commissures. Since the serological test for syphilis was never positive and the aortic lesion was atypical in several features, a relationship to the "collagen diseases" may be suspected.

The characteristic lesion of the "collagen diseases" is fibrinoid necrosis of connective tissue. Klinge,[8] introducing the term, believed the change to be the result of swelling and chemical alteration of the ground substance. Altshuler and Angevine [9] have recently considered fibrinoid necrosis to be marked by a precipitation of "acid muco-polysaccharides" in altered collagen.

The carbohydrate content of glomeruli and of the hyalin accumulating in obsolescent glomeruli assumes importance in view of the carbohydrate of fibrinoid necrosis. It is possible that the carbohydrate in the collagen may be the cause of the necrosis rather than its result. The concept that antigenicity is a property solely of protein has been refuted thoroughly by the demonstration that polysaccharides are antigens in the case of pneumococci, blood group substances and so on. The ground-substance carbohydrate or some slightly altered component of it may actually be the antigen setting off the sensitivity phenomena of the collagen diseases.

It is possible that the obsolescent glomeruli in the present case are antigenic by virtue of altered carbohydrate. One case in which obsolescent glomeruli were the seat of intense lymphocytic reaction in a case of chronic glomerulonephritis in an infant has been described and illustrated.[2] The case under discussion presents a similar appearance with the difference that macrophages forming tubercles are characteristic rather than lymphocytes. Either or both may represent antibody-antigen activity about altered glomeruli.

The possibility of granuloma formation being interpreted as an immunity phenomenon has been reviewed by Forbus.[10] It is pointed out that "with few exceptions, those diseases in which outstanding immunity develops as a result of infection are the diseases in which the granulomatous inflammatory reaction is most conspicuous." Forbus is "not sure that the evidence (for granulomas being associated with antibody formation) is more than suggestive." The collagen diseases have been considered as examples of hypersensitivity, most recently from the studies of Rich and Gregory.[11] The present case may be important in this regard as combining granuloma formation with the other features of collagen disease.

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7. Libman, E., and Sacks, B.: A Hitherto Undescribed Form of Valvular and Mural Endocarditis, Arch. Int. Med. **33**:701-737 (June) 1924.

8. Klinge, F.: Der Rheumatismus, Engebn. allg. u. path. Anat. **27**:1-351, 1933.

9. Altshuler, C. H., and Angevine, D. M.: Histochemical Studies on the Pathogenesis of Fibrinoid, Am. J. Path. **25**:1061-1077, 1949.

10. Forbus, W. D.: Granulomatous Inflammation, Springfield, Ill., Charles C Thomas, Publisher, 1949.

11. Rich, A. R., and Gregory, J. E.: The Experimental Demonstration That Periarteritis Nodosa is a Manifestation of Hypersensitivity, Bull. Johns Hopkins Hosp. **72**:65-88, 1943.

There is not much doubt that the increasing number of cases of "collagen diseases" being recognized by pathologists represents a real increase in their frequency as well as a popularizing of the concept. In these circumstances reports of unusual cases seem justified when a peculiar and apparently new appearance is described.

SUMMARY

A case in which an unusual granulomatous lesion of renal glomeruli was demonstrated is presented. This lesion existed with clinical manifestations of polyarthritis. Microscopic evidence of an acute necrotizing arteritis was also present.

This case has been compared with previously reported cases in which glomerulonephritis and arthritis have been associated. Similarities between the findings and those described in the "collagen diseases" have been discussed.

University of Virginia School of Medicine, Charlottesville (Dr. McManus).

Duke University School of Medicine, Durham, N. C. (Dr. Hornsby).

PORTAL AXIS THROMBOSIS WITH SPONTANEOUS PORTACAVAL SHUNT AND RESULTANT COR PULMONALE

LIEUTENANT COLONEL FRANK A. MANTZ Jr.
MEDICAL CORPS, UNITED STATES ARMY
AND
ERNEST CRAIGE, M.D.
BOSTON

CHRONIC pulmonary hypertension and chronic portal hypertension are well recognized by clinicians and pathologists as distinct clinical entities, ordinarily of separate cause and pathogenesis. A case in which the two are coexistent and interrelated is unique. Such a case forms the basis of this report. In this case a shunt between the portal system and the vena cava formed a devious path through which multiple emboli passed to the lungs, resulting in pulmonary hypertension, chronic cor pulmonale and death.

HISTORY OF CASE

A widow (M. G. H. 663390), 53 yr., was referred to the Massachusetts General Hospital in May, 1949, because of hoarseness of 18 months' duration. She gave a past history of four episodes of massive hematemesis between 1929 and 1938. Treatment of the last of these included 10 transfusions. Roentgenograms in 1938 showed "no evidence of pulmonary pathology, but . . . tortuous defects extended up and down the entire esophagus with a roentgenographic appearance of varicose veins . . ."

Since 1938 she had been well, noticing only mild dyspnea on hills. In Feb. 1949, there was sudden onset of hoarseness. Because of this, she came to the Out-Patient Department of the Massachusetts General Hospital in May, 1949.

Physical examination disclosed a well developed and well nourished middle-aged woman with no cyanosis or clubbing. The heart was enlarged, with an apical impulse 12 cm. from the midsternal line. The pulmonary second sound was duplicated and accentuated. No murmurs were heard, and there was a gallop rhythm. The lungs were clear. The blood pressure was 117 systolic and 72 diastolic. The pulse rate was 85. Examination of the abdomen revealed no abnormality. There was minimal edema of the ankles. The hemoglobin content was 13.7 gm. Roentgenograms showed the heart to be enlarged. The pulmonary arteries were prominent. A barium sulfate swallow showed irregular defects in the esophagus, but its walls were not fixed (Fig. 1). Esophagoscopy showed the left vocal cord to be completely paralyzed. The esophageal mucosa was thrown into folds by extrinsic pressure. The mucosa was normal, and no dilated veins were seen. A biopsy was noncontributory. An electrocardiogram showed the pattern of right ventricular hypertrophy.

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Research fellow in pathology, Massachusetts General Hospital (Lt. Col. Mantz); assistant in medicine, Harvard Medical School, and assistant in medicine, Massachusetts General Hospital, (Dr. Craig), with the aid of the National Heart Institute, United States Public Health Service.

On Sept. 14, 1949, she was admitted to the surgical service and an exploratory thoracotomy was performed, since the possibility of a mediastinal tumor could not be excluded. The pulmonary artery was tremendous, and it had pulsations simulating those of the aorta. A huge periesophageal plexus of veins was discovered. No tumor could be demonstrated. The patient's pulse and blood pressure were poor during the operation. The chest was closed quickly, and she was given 2,000 cc. of whole blood. Hypotension persisted, and she became slightly cyanotic, with signs also of right-sided heart failure. Death occurred on the fourth postoperative day.

NECROPSY

Necropsy (performed 13 hr. post mortem) disclosed a remarkable portacaval shunt originating at the superior aspect of the confluence of the portal, the splenic, and the mesenteric vein, hereinafter referred to as the portal axis. This extended upward beneath the gastrohepatic ligament along the lesser curvature of the stomach, from which it received numerous dilated and engorged contributory veins. The

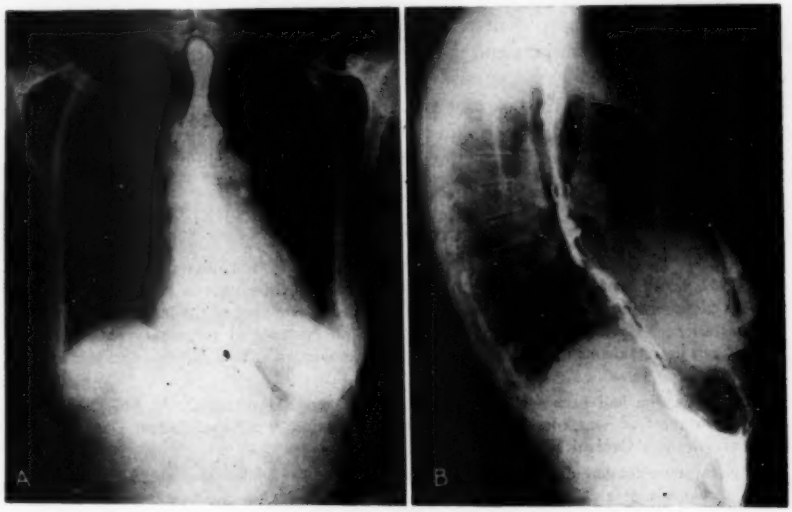

Fig. 1.—*A*, posterior-anterior roentgenogram of the chest showing prominent pulmonary arteries and the tortuous course of the barium sulfate-filled esophagus. *B*, lateral view of the chest.

abnormal vessel passed through the esophageal hiatus and coursed along the posterior aspect of the esophagus as a dilated cirsoid varicose channel. It varied from 1.5 to 3 cm. in diameter and its greatest bulk was found on the left posterolateral aspect of the esophagus, where it lay in intimate relationship with the left recurrent laryngeal nerve. At the level of the cricoid cartilage the vein curved anteriorly and downward along the right side of the esophagus to enter the right innominate vein apparently as an independent vessel (Figs. 2 and 3). It did not communicate with the azygos or hemiazygos system at any point. The walls of the abnormal channel were thickened, fibrous, and focally calcified. Varying amounts of gray-brown thrombus in different stages of organization were adherent to the lining in the majority of the varicose pockets.

The portal axis was converted into an irregular venous sinus about 4 cm. in diameter with extremely fibrous and calcified walls. The intimal aspects were roughened by large amounts of adherent organized thrombus, but the orifices of the various channels opening into this chamber were not significantly reduced in caliber. The surrounding stroma was exceedingly dense, forming a scarlike cushion in which the portal axis was embedded.

Fig. 2.—Liver, spleen, celiac axis, and portal system showing varicose portacaval shunt emptying into descending vena cava through dilated innominate vein.

The superior mesenteric and splenic veins likewise showed numerous varicose dilatations and followed tortuous courses. These passages presented thickened, partially calcified, fibrous walls lined irregularly with gray tan, laminated, organized thrombi. The inferior mesenteric vein was dilated but otherwise unremarkable.

The esophageal wall was edematous and contained numerous thin-walled venous varicosities which communicated with the paraesophageal vessel described above. These were most prevalent in the distal third, where they frequently projected into the lumen, covered only by a thin layer of mucous membrane.

The serosal veins of the stomach were unusually prominent, particularly on the lesser curvature aspect. The intestines were hyperemic.

The peritoneal cavity was free of exudate and adhesions. The liver was of average size and weighed 1,030 gm. The parenchyma showed well preserved lobular markings, with no evidence of infarction. No vascular lesions were found within the liver, but the portal vein was strikingly narrowed to about one-fifth its normal size, measuring only 5 mm. in diameter. The wall was thickened and fibrous, with occasional small subintimal atheromatous deposits. No valves were apparent, and no thrombi or other obstructive lesions were discovered on gross dissection into the major branches.

Approximately 100 cc. of clotted blood was present in the left pleural cavity. The lungs together weighed 1,050 gm. Both lungs exhibited moderate passive

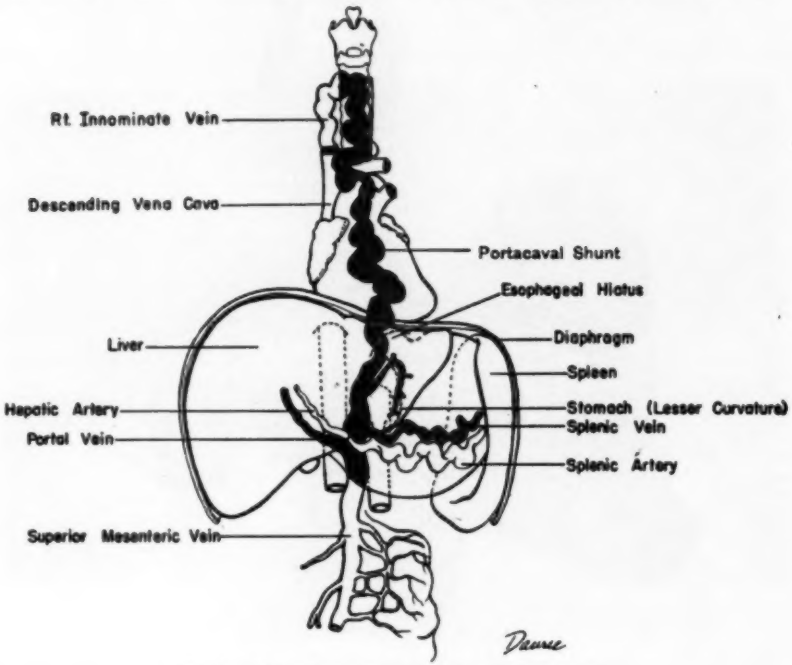

Fig. 3.—Schematic representation of portacaval shunt.

congestion and edema, and the left lung was moderately atelectatic. No pulmonary infarcts were found. The pulmonary artery with its major branches was markedly dilated, measuring 12.5 cm. in circumference at the conus. Numerous bright yellow deposits of atheromatous material were present in the intima of the conus and its branches as far out as the third and fourth order. No emboli or thrombi were demonstrated grossly. The bronchial tree and pulmonary veins were not remarkable.

The heart weighed 300 gm. and showed slight right-sided preponderance with a cardiothoracic ratio of 16:23 cm. The right ventricular wall measured 7 mm. in thickness and showed striking hypertrophy of its columnae carneae and papillary muscles. The left ventricular wall measured 14 mm. in thickness and was not

abnormal. Both atria and the right ventricular chamber were moderately dilated. There was no evidence of endocardial or myocardial disease, and no mural thrombi were present. The mitral valve measured 11.5 cm. in circumference; the aortic valve, 8 cm.; the tricuspid valve, 15 cm., and the pulmonic valve, 8.7 cm.

The spleen was enlarged, weighing 500 gm. Its capsule was thickened and fibrous. When the organ was sectioned, there was gritty resistance, and large amounts of blood were released from the cut surfaces. The trabeculae were widened, but the follicles were obscured. Several dilated veins gave a honeycomb appearance to the cut surfaces adjacent to the hilus, and many of these contained calcium within their walls. The splenic artery was exceedingly enlarged, sclerotic, partially calcified, and presented numerous cirsoid aneurysmal dilatations extending from the hilus to

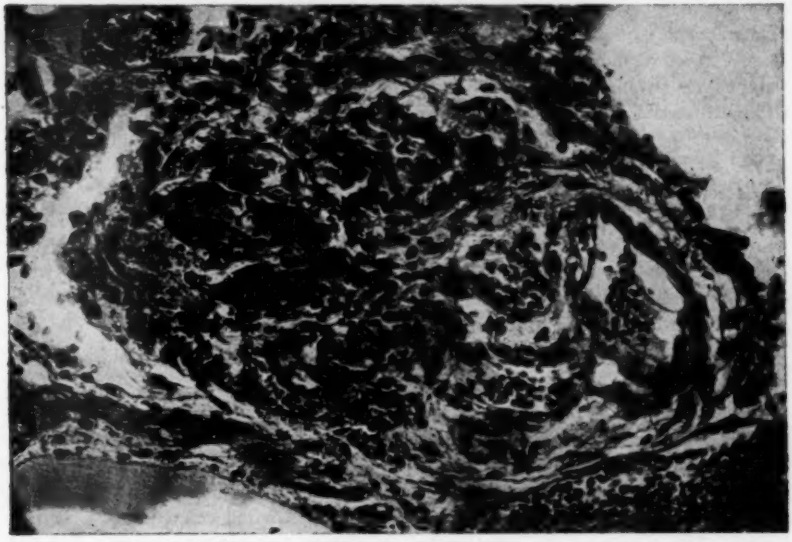

Fig. 4.—Recently embolized small pulmonary artery in which there has been marked endothelial proliferation, early organization, and recanalization.

the celiac axis. No anastomoses could be demonstrated between the splenic artery and the varicose splenic vein.

The remaining organs were free of noteworthy abnormalities. No thrombi were present in the femoral, the saphenous, or the iliac veins.

MICROSCOPIC EXAMINATION

Sections from all lobes of both lungs revealed embolization of numerous terminal arteries and arterioles by amorphous eosinophilic fibrinous material consistent in appearance with old blood clot. Various stages of organization and recanalization were observed from early intimal proliferation to complete scarring (Fig. 4). The early lesions were interesting in that they showed extreme endothelial proliferation producing a glomerular appearance within the lumen. The older lesions showed

moderate medial and intimal proliferation and fibrosis, but no associated inflammatory reaction was observed. Rarely a completely scarred terminal artery was seen. Many arteries of the next higher order showed hyperplastic sclerosis. The medium-sized and large pulmonary arteries displayed moderate to marked intimal thickening with hyalinization and atherosclerosis. The media of several of these vessels was hypertrophied and exhibited variable edema with increase in basophilic mucoid ground substance. The appearance of the pulmonary parenchyma was not remarkable.

Examination of the heart did not show significant changes. Sections from the portal, splenic and superior mesenteric veins at the site of their confluence and from the portacaval shunt were essentially similar. Their inner lining was thickened irregularly and composed of laminated, hyalinized, faintly basophilic material containing widely spaced pyknotic, spindle-shaped cells in parallel arrangement. The appearance of lamination was enhanced by the presence of occasional cleftlike spaces sometimes containing erythrocytes. Focal areas of calcification were found within the hyaline stroma. Elastic tissue stains confirmed the intimal position of the hyaline material and revealed irregular accumulations of elastic fibers within this substance, suggesting attempts at recanalization of a thrombus. The outer portions of the walls were exceedingly thin and composed of atrophic bundles of smooth muscle in loose arrangement.

The splenic pulp revealed slightly increased cellularity, but no abnormal cells were seen. The sinusoids were surprisingly collapsed and thin-walled. The capsule and the trabeculae were markedly fibrous. The branches of the splenic vein were dilated conspicuously and thick-walled. A large parenchymal branch of the splenic artery was exceedingly sclerotic and partially calcified.

Examination of the remaining organs did not contribute additional significant information.

COMMENT

The portacaval shunt discovered in this case followed a most unusual pattern, and the preponderance of the paraesophageal vein noted is decidedly rare. Simonds [1] in an extensive review of portal vein occlusion did not record a single anastomosis of this type, and we have been unable to find reference to a similar lesion. The course of the anastomotic channel in our patient suggests that the coronary, the paraesophageal, and probably the inferior thyroid veins contributed to its continuity. The failure to discharge the majority of its contents into the azygos system probably accounts for the extreme size and upward extension of the shunting vessel. It is obvious that the dilated paraesophageal channel and its numerous communications with the submucosal veins of the esophagus produced the alterations noted on roentgenologic examination and most certainly explained the widely spaced hematemeses experienced by the patient. The paralysis of the left recurrent laryngeal nerve may have resulted from compression of the nerve either by the varicose venous mass or by the widely dilated left pulmonary artery.

The pathogenesis of this abnormality is not completely clear. The fact that in the history there was no event compatible with sudden acute disease of the upper abdominal region would seem to preclude the usual rapid development of portal

1. Simonds, J. P.: Chronic Occlusion of the Portal Vein, Arch. Surg. **33**:397-424 (Sept.) 1936.

thrombophlebitis and serves to exclude acute primary disease of the liver or adjacent organs. The high degree of stenosis within the portal vein, on the other hand, suggests primary phlebothrombosis many years before, and it seems more likely that the thrombosis was gradual in development and never complete. The possibility of preexisting congenital hypoplasia of the portal vein cannot be excluded, and its exceedingly narrow caliber strongly suggests that such a factor may have prevailed.

It is apparent from the history that mild cardiac embarrassment, manifested by dyspnea on exertion, existed during the last 10 yr. of life. Fully 5 mo. before death the patient's physical findings strongly suggested pulmonary hypertension. The pulmonary changes discovered at autopsy were those of prolonged increase in intravascular tension, as evidenced by the diffuse hyperplastic sclerosis of the small and medium branches of the pulmonary artery, the severe atherosclerosis of the larger branches and the hypertrophy of the right ventricle.[2] The causes of pulmonary hypertension and sclerosis are diverse and known to be both intrapulmonary and extrapulmonary. In this instance there seems little doubt that multiple pulmonary embolizations were the precipitating factors. The role of such lesions as a cause of cor pulmonale has recently been emphasized by Castleman and Bland.[3] The conclusion that the vascular lesions in the present case represent embolization rather than thrombosis rests solely on the obvious source of emboli within the portacaval anastomosis and the lack of factors conducive to long-standing pulmonary thrombosis within the lungs. The confinement of small emboli to scattered pulmonary arterioles probably accounts for the lack of infarction. It may be assumed, in the absence of pulmonary passive congestion, that sufficient collateral circulation was available through the bronchial arteries to prevent ischemic necrosis of tissue.

SUMMARY

An unusual instance of a spontaneous portacaval shunt of undetermined origin (? congenital) is recorded. The anastomotic channel gave rise to multiple pulmonary emboli, widespread obstruction in the vascular bed of the lungs, pulmonary hypertension, chronic cor pulmonale and death.

2. Brenner, O.: Pathology of the Vessels of the Pulmonary Circulation, Arch. Int. Med. **56**:211-237 (Aug.); 457-497 (Sept.); 724-752 (Oct.); 976-1013 (Nov.); 1189-1241 (Dec.) 1935.

3. Castleman, B., and Bland, E. F.: Organized Emboli of the Tertiary Pulmonary Arteries, Arch. Path. **43**:581-589 (Dec.) 1946.

A NOTE ON THE ORIGIN OF MULTINUCLEATED GIANT CELLS FROM VASCULAR CHANNELS IN TUMORS

Tumors Arising in Thyroid Gland, Bone, and Soft Tissue

LELLAND J. RATHER, M.D.
SAN FRANCISCO

THE PURPOSE of this communication is to point out the apparent origin, from the inner lining of vascular channels, of multinucleated giant cells forming a striking feature of a rare malignant tumor of the thyroid and of the common giant cell tumor of bone, and to comment on their occurrence in other lesions. The thyroid tumor referred to has been reported in detail elsewhere,[1] and a brief summary will be sufficient for the purpose of the present paper.

SUMMARY OF DATA ON TWO CASES

CASE 1.—The patient, a white woman aged 55, complained of hoarseness and unilateral swelling in the thyroid region of about six weeks' duration. At operation a hard mass was found to involve the left upper part of the thyroid, extending across the midline. Complete removal was not possible, but 75 Gm. of firm pink fleshy tissue was excised. Histologically, this consisted of a dense, matlike stroma of reticular and collagen fibrils and two cellular elements, the first comprising spindle cells with poorly defined eosinophilic cytoplasm and nuclei of highly variable size, contour, and staining quality; the second comprising the characteristic multinucleated giant cells to be described. The nuclei of the former were predominantly oval in outline, but some were very large (up to 30 μ) hyperchromatic, and of irregular outline. The multinucleated cells were distributed more or less uniformly throughout and averaged 90 μ in diameter, although some were much larger. They contained up to a hundred or more centrally located nuclei in the plane of the sections, most of the nuclei being uniform in size, shape, and staining characteristics, round or oval, about 8 μ in diameter, with diffusely distributed chromatin, and containing neither mitotic figures nor prominent nucleoli. The cytoplasm was abundant and eosinophilic. Many of them lay in thin-walled vascular channels filled with intact red cells and it appeared that they took origin from the inner lining cells (figs. 1 and 2). The patient was treated postoperatively with a total dose of 4,000 r (air). She died at home three months after the operation, and an autopsy was not obtained.

The giant cell tumor of bone in which similar multinucleated cells will be shown has not been reported heretofore.

CASE 2.—A white woman aged 26 presented herself with the complaint of persistent pain in the right knee, of four months' duration. Roentgenograms showed in the lower part of the right femur a destructive lesion which was thought to be most probably a metastatic tumor. At operation 60 Gm. of friable, coarsely granular, reddish brown material was removed. The histological diagnosis was benign giant cell tumor. Nine months later a recurrence was removed from beneath the skin at the operative site. The diagnosis was again benign giant cell tumor, but

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1. Rather, L. J.: Giant Cell Tumors of the Thyroid, Stanford M. Bull. 8:202-208 (Nov.) 1950.

the presence of fairly numerous mitotic figures and variability of size of the "stromal" cell nuclei were noted. Multinucleated cells similar to those seen in the thyroid tumor were present in relation to vascular channels (figs. 3*A* and 4).

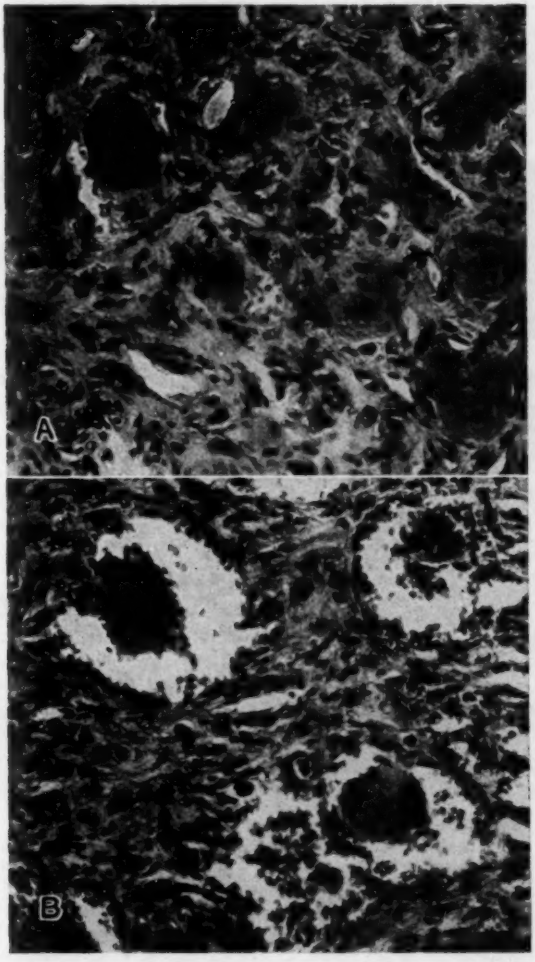

Fig. 1 (case 1).—Giant cell tumor of the thyroid; hematoxylin and eosin stain; × 400. *A* shows multinucleated cells with intracytoplasmic channels. Note the nuclear variation of "stromal" cells. *B* shows multinucleated cells projecting into vascular channels containing intact erythrocytes.

COMMENT

The occurrence in the thyroid gland of a tumor bearing a close resemblance to giant cell tumor of bone is uncommon but has been reported. Wegelin,[2] after dis-

2. Wegelin, C.: Handbuch der speziellen Anatomie und Pathologie, Berlin, Julius Springer, 1926, vol. 8, p. 288.

cussing pleomorphic carcinoma of the thyroid with tumor giant cells, referred to a rare thyroid tumor with multinucleated cells resembling osteoclasts scattered among smaller anaplastic cells. Nadal [3] reported such a case in 1910. The patient, a woman aged 48, noticed a sudden and rapid enlargement of a goiter which she had had for many years. An exploratory operation revealed a tumor which replaced the right lobe and extended across the isthmus. Complete removal was not possible, and the patient survived for only a few months after the operation. The characteristic histological feature was the presence of multinucleated giant cells with 70 or

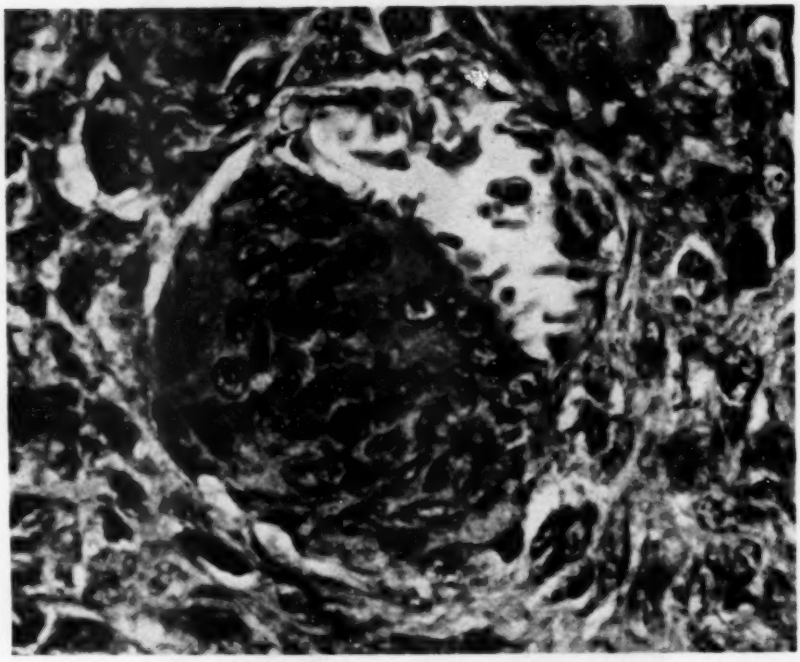

Fig. 2 (case 1).—Giant cell tumor of the thyroid. The vascular channel contains intact erythrocytes. The cytoplasmic margin of the multinucleated giant cell is continuous with the channel wall. Hematoxylin and eosin stain; × 1300; Zeiss phase contrast photomicrograph made by A. T. Brice, Ross, Calif.

more small, oval, centrally disposed nuclei. These lay among spindle-shaped cells which did not appear to be of epithelial origin. Nadal stated that this was indistinguishable from giant cell tumor of bone (*sarcome à myéloplaxes*) and pointed out that the presence in the thyroid of a tumor type considered proper to bone was difficult to explain. He suggested, as alternative possibilities, that the resemblance was superficial and fortuitous, that the giant cell tumor of bone did not originate from medullary elements as had been thought, or that the lesion was comparable to the mixed tumor of the parotid. Just what he meant by the last statement is not

3. Nadal, P.: Sarcome à myéloplaxes, Bull. Soc. anat. de Paris **85**:736-737, 1910.

clear; perhaps he may have had carcinosarcoma in mind. As for the similarity of the lesion to giant cell tumor of bone, it is probable that Nadal did not put enough

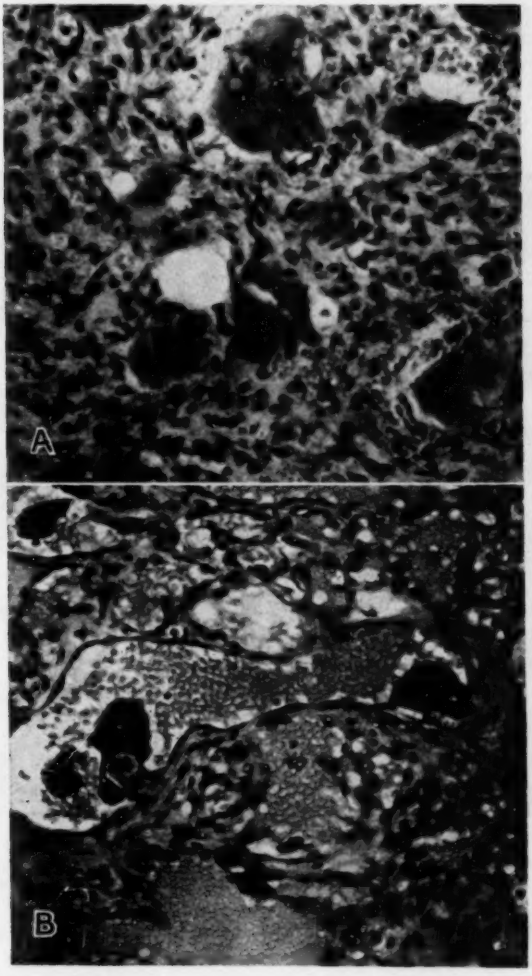

Fig. 3 (case 2).—*A*, giant cell tumor of bone showing multinucleated cells with intracytoplasmic channels. The "stromal" nuclei are fairly uniform. Hematoxylin and eosin stain; × 400. *B*, so-called pyogenic granuloma of skin, showing multinucleated giant cells projecting into vascular channels. The "stromal" cells are uniform. Hematoxylin and eosin stain; × 400.

emphasis on the characteristics of the "stromal" cells, since it is likely that in his case, as in ours, anaplasia of these cells was more pronounced than in giant cell tumor of bone (except in rare instances).

The resemblance of these tumors of thyroid and bone is, however, more than superficial, since in the two lesions the multinucleated giant cells seem to be of similar origin. Johnson [4] has summarized the literature relating to these characteristic cells of giant cell tumor of bone, and reference to his paper may be had for a fuller exposition. Briefly, Virchow (1864) and Borst (1897) believed that they were hypertrophied bone cells set free by the absorption of bone matrix and identical with osteoblasts; Wegner (1878) thought them related to the endothelium of angioblastic tissue, while Stroebe (1890) stated flatly that they developed from

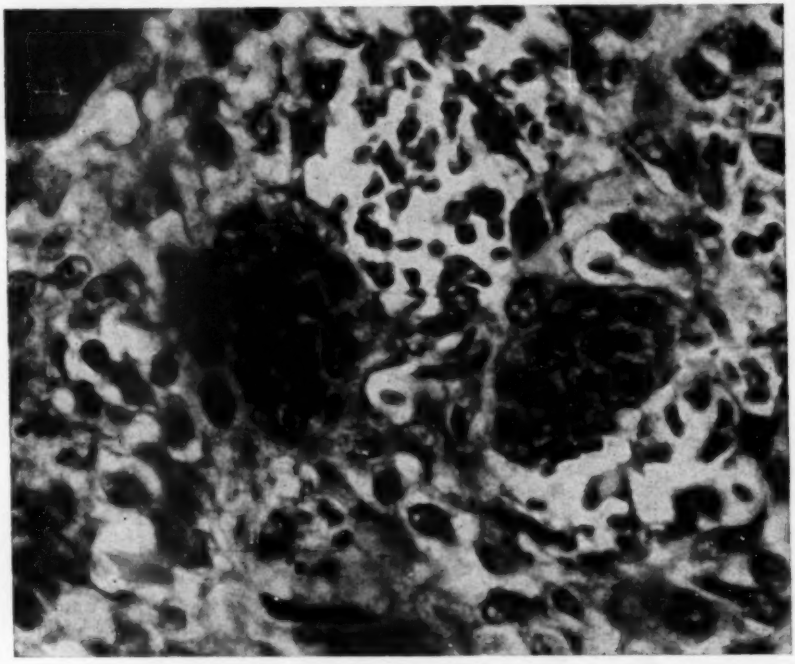

Fig. 4 (case 2).—Giant cell tumor of bone. That the cytoplasmic margin of the multinucleated cells is continuous with the wall of the channel is clearly shown. Hematoxylin and eosin stain; × 1300; Zeiss phase contrast photomicrograph made by A. T. Brice, Ross, Calif.

endothelial cells, to which Lubarsch (1895) agreed. Mallory (1911) proposed that they were foreign body giant cells formed by the fusion of large mononuclear phagocytes. Aegerter [5] reviewed the problem recently and presented the belief that the multinucleated giant cells found in a number of bone lesions (including giant cell tumor of bone, giant cell tumor of tendon, hyperparathyroidism, fibrous dysplasia, Paget's disease) were identical, or generically related, and produced for the

4. Johnson, W. W.: The Giant Cells of Benign Giant Cell Tumors of Bone, Arch. Path. **10**:197-205 (Aug.) 1930.

5. Aegerter, E. A.: Giant Cell Tumor of Bone, Am. J. Path. **23**:283-293 (March) 1947.

purpose of phagocytosis, as are the osteoclasts. This is not in agreement with the reported finding [6] that osteoclasts do not behave as macrophages in disintegrating bone and do not contain calcium, as do ordinary macrophages under similar circumstances, nor is it in agreement with the conception of the histogenesis of the multinucleated cells in the lesions under discussion from vascular channels.

Johnson carried out a detailed examination of 11 giant cell tumors of bone, from the files of the Stanford Medical School Department of Pathology, and found that the giant cells occurred most commonly in association with intact red blood cells, without evidence of extravasation in the form of significant amounts of pigment or fibrosis. In all the specimens he found the giant cells to line, or lie in relation to, thin-walled vascular channels. He gave convincing illustrations of his contention that the giant cell cytoplasm extended into the endothelial wall and that the cells thus directly lined the channel. He confirmed the older observation of Stroebe that blood channels could be seen within the cytoplasm of the giant cells, a finding usually interpreted as intracellular vacuolation. This relation of the multinucleated giant cells to vascular channels containing intact erythrocytes is shown in the accompanying photomicrographs. Intracellular vascular channels are best seen in figures 1 *A* (the thyroid tumor) and 3 *A* (the bone tumor). In figures 2 and 4, taken with a Zeiss phase contrast microscope, the continuity of the thin vascular wall with the cytoplasmic border of the giant cells is visible.

Since this observation came to our attention my associates and I have seen a similar disposition of multinucleated cells in other giant cell tumors of bone, in giant cell tumor of tendon, and in several examples of epulis. The histogenesis of the benign giant cell tumor of tendon has been restudied recently by Foster,[7] who concluded that this lesion is an example of sclerosing hemangioma whose multinucleated cells are formed by the coalescence of endothelial cells. The manner of formation of these cells, according to the formulation advanced in the present paper, is most clearly shown in figure 4 from an example of the so-called pyogenic granuloma of the skin.

SUMMARY AND CONCLUSIONS

The significance of these findings seems to be that multinucleated giant cells may originate as reactive formations in blood vascular channels in a variety of otherwise unrelated lesions, including tumors of the thyroid, bone and soft tissues. In some tumors of angioblastic nature the multinucleated cells are actually part of the tumor proper. Whether occurring in benign or in malignant tumors, or in nonneoplastic lesions, their site of origin may be the same. Because of their nonspecificity of occurrence these multinucleated giant cells are unsuited for the classification of such lesions.

2398 Sacramento St.

6. McLean, F. C., and Bloom, W.: Calcification and Ossification, Arch. Path. **32**:315-333 (Sept.) 1941.

7. Foster, L. N.: The Benign Giant Cell Tumor of Tendon Sheaths, Am. J. Path. **23**:567-576 (July) 1947.

Notes and News

Third Annual Lecture Honoring Dr. S. Burt Wolbach.—The Third Annual Lecture Honoring Dr. S. Burt Wolbach, Emeritus Shattuck professor of pathological anatomy, Harvard Medical School, was delivered on May 14, 1951, at the Peter Bent Brigham Hospital, Boston, by Dr. George M. Haas, director of laboratories, Presbyterian Hospital and professor of pathology (Rush) College of Medicine, University of Illinois. The title of the lecture is "Isolation and Characterization of Tissue Structure."

Portrait of Dr. Whipple.—A portrait of Dr. Whipple, dean of the University of Rochester School of Medicine and Dentistry, has been presented to the Rochester Academy of Medicine as a gift of its past presidents.

Award.—Dr. Fred W. Stewart, pathologist for the Memorial Hospital Center for Cancer and Allied Diseases, New York, was the recipient of the 1951 annual Bertner Foundation Award, and delivered the Bertner Lecture at the University of Texas M.D. Anderson Hospital's Fifth Annual Symposium on Fundamental Cancer Research at the Shamrock in Houston on April 21. Established in 1950, the Bertner Award and Lectureship is given each year to an individual selected for his outstanding contributions to the field of cancer research.

Appointments.—On July 1, 1951, Dr. Robert E. Stowell of the University of Kansas Medical Center, Kansas City, presently chairman of the department of oncology will become chairman of the department of pathology, and a combination of the two departments will be effected. This reorganization follows, the retirement of Dr. H. R. Wahl, who has served as professor of pathology and chairman of the department.

Dr. Orville Bailey has been appointed professor of neuropathology at Indiana University School of Medicine. Formerly Dr. Bailey was assistant professor of pathology at Harvard Medical School and neuropathologist and vice-chairman of the Neurological Institute at the Childrens' Medical Center, Boston.

American Board of Pathology.—The next examination of the American Board of Pathology will be held at Northwestern University Medical School, Chicago, on Oct. 11, 12, and 13, 1951. All correspondence related to this should be addressed to the newly elected secretary-treasurer, William B. Wartman, M.D., Department of Pathology, Northwestern University Medical School, 303 E. Chicago Ave., Chicago 11. Dr. Wartman succeeds Dr. Robert A. Moore as secretary-treasurer of the board.

Deaths.—Henry Montgomery Weeter, former professor of bacteriology at the Louisville University School of Medicine died February 25, aged 63, of coronary infarction. Dr. Weeter was a specialist certified by the American Board of Pathology; a member of the College of American Pathologists and the American Society of Clinical Pathologists; past president of the Jefferson County Medical Society and the Kentucky Society of Pathologists.

Dr. Ruell A. Sloan of Chevy Chase, Md., and presently curator of the Army Medical Museum, Washington, D. C., died suddenly at his home on Sunday, June 17, 1951. He was 42 years of age. Dr. Sloan received his M.D. degree from the University of Rochester in 1935. He had held teaching appointments at Harvard Medical School, St. Louis University School of Medicine, and the University of Buffalo School of Medicine. During World War II he served as executive officer of the Army Institute of Pathology, and in 1947 was appointed curator of the Army Medical Museum. Dr. Sloan was a member of numerous medical societies and during the past several years he has served ably as the secretary-treasurer of the International Association of Medical Museums and of the American and Canadian Section of this association.

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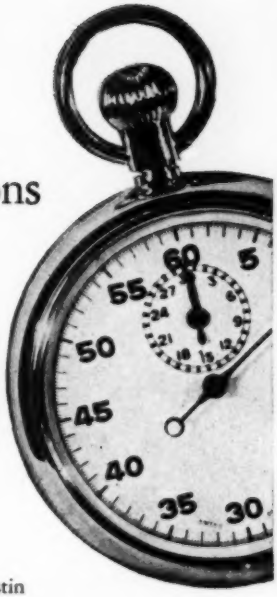

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1. Shapiro, S., Weiner, M., et al.: Am. Heart J. **40**: 766 (Nov.) 1950

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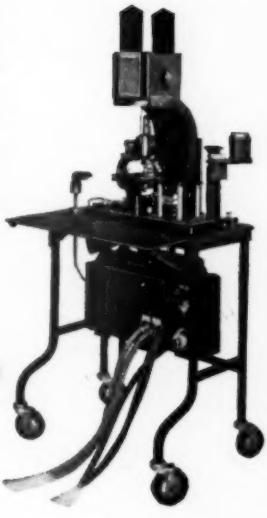

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